nature

19 February 1981

Employment, training and opportunity

Economic recessions, painful though they may be, are supposed in the conventional wisdom also to be opportunities for industrial change. Industries whose competitiveness has been undermined close factories, throw workers into the dole queues and perhaps go out of business. In due course, the theory goes, some of the people thus displaced from work become the entrepreneurs who found new kinds of industries; others learn new skills so that when the economy revives they too can join the workforce in a new pattern of industry. Adversity, the textbooks say, can thus be an instrument of rapid change. So does it follow that the wave of recession now sweeping the industrialized West is merely a sign of better things to come? Not if Britain is an example, or if the British government's capacity to make the best of the opportunities now presented is typified by its shabby performance in the debate on industrial training in the House of Commons last week.

Nobody disputes the severity of the recession now afflicting the British economy. Manufacturing output has declined by some 15 per cent since its peak two years ago. Employment in the private sector has declined even faster (which promises increased productivity when the worst is over). Unemployment now exceeds 2.5 million, is most serious among young people and threatens to go on increasing at least until the summer. So is this not a splendid opportunity for making sure that the people out of work are well prepared for the recovery that may yet come? Or at least for giving them incentives to acquire new skills if they are so inclined? Both the government and the Labour opposition seem to agree, if last week's debate is to be taken seriously. Their good intentions are not matched by an imaginative appraisal of what should be done.

The narrow issue that divides them is the future of the network of industrial training boards, first set up in 1964 and financed by means of a levy on the turnover of all companies operating in specified groups of industries. The system is not as onerous as it may seem. Intelligent companies ensure that they recover at least as much in financial support for their internal training operations as they pay by way of levy. Some of the best apprentice schools in Britain are now financed by means of this genteel laundering of money, and the system may also blunt the edge of the longstanding discontent of progressive companies that much of the training they provide eventually benefits less farsighted competitors which pick up skilled men in the job market. The system of training boards has probably also served a useful purpose in industries such as catering, where individual companies tend to be small and where the labour force is mobile. The system, nevertheless, has serious weaknesses. From the start, the training boards have been inclined to impose on the industries they supposedly serve patterns of training whose utility is disputable. Since 1973, when the administration costs of the training boards became a charge on the central government, the boards have become even more bureaucratic than they were before, and extravagant to boot. (At least one director of a training board is paid more than a university vice-chancellor at a British university.)

For several months, it has been clear that this state of affairs could not continue. Even the agencies of the British government sympathetic to the training boards (the Manpower Services Commission, for example) have acknowledged that the system has serious faults. Industrial companies, especially progressive companies, have become increasingly discontented with the arrangements. A few months ago, the Conservative Party's Centre for Policy Studies advocated the simple abolition of the training boards. The Conservative government, as much concerned with the cost of keeping the boards in being (more than £50 million a year) as with the need for training, has resolved to transfer the cost of administration back to the industries which contribute to the boards. The case for this course of action, put forward in the House of Commons last week, is that a transfer of full responsibility for the boards back to the industries concerned would provide a built-in assurance that industrial training is geared to industrial needs.

The argument, so far as it goes, makes sense. It is, however, disingenuous to suppose that there will be no consequential changes. Many industries now saddled, as they see it, with training boards will regard the Employment and Training Bill now working its way through the House of Commons as a licence to do away with their training boards. In many cases, the result would be far from tragic. Many of the complaints recently levelled at the boards are fair. One serious weakness is that there has never been an objective evaluation of the educational value of their training schemes. A more serious weakness is structural. If the best outcome of the present recession of the British economy is the emergence of entirely novel industries, what purpose can be served in perpetuating a pattern of training geared to industries in decline? Although true, none of it excuses the narrowness of the British government's case for change.

Everybody accepts that this point in the current business cycle calls for new initiatives in training. The British government's argument that the responsibility should as a matter of principle fall squarely on industry is, however, profoundly flawed. Progressive industrial companies have always acknowledged that their own self-interest requires some investment in the skills of their own workpeople, and will continue to do so. Most of them acknowledge that the cost is worthwhile, even if some of their skilled people eventually work for competing companies. But the same industrial companies have no interest in providing their workpeople with skills suited to entirely novel industries. The result is that people working in, say, traditional mechanical engineering are unlikely to be given the chance, within their own organizations, of becoming skilled in electronic engineering or computing. And what of those who are out of work, and thus outside the scope of the industrial training boards? In Britain, in the past few years, the Manpower Services Commission has done some useful work in keeping young people off the streets and providing them with modest skills, but none of this amounts to the creation of the body of skill on which an entirely novel pattern of industry might be built some years from now.

One paradox is that a government devoted, so far as can be told from its public declarations, to individual initiative should so overlook what is now the obvious need — that people by their individual decisions should more easily be able to acquire the skills they consider would help them to make their way in the brave new world ahead. Another paradox is that there is no shortage of facilities for the provision of training, and, by all accounts, no shortage of people seeking skills. Both the universities and the polytechnics are at present short of students and, faced with declining support for conventional students' education, more eager than ever to provide vocational training of various kinds. So why does not the potential demand for advanced training more fully meet the potential supply? At least one part of the explanation is the cost. Those in jobs must pay the full cost of a training course on which they embark out of their taxed income. Those out of jobs are usually unable to raise the

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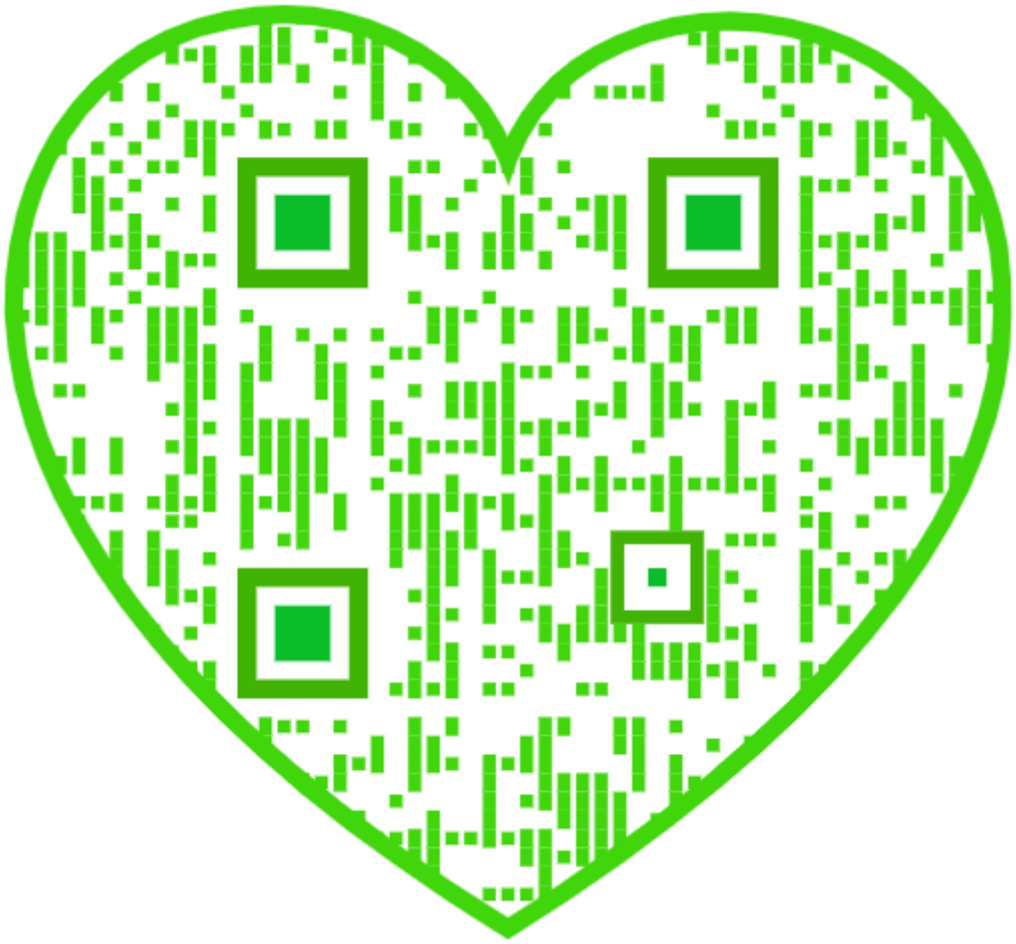

fees at all. The implications are plain. If the British government means what it says about the importance of training for the future health of British industry, it must make the cost of training fees paid out of a taxpayer's pocket tax-deductible. And it must use some of the £50 million a year it will save on the industrial training boards to help those out of work to acquire new skills.

In the past two years, the British government has been outwardly resolute in sticking to the monetarist guns that have made the present recession the painful phenomenon it is. The government has been alternatively praised for courage and pilloried for pigheadedness. In reality, it has followed an intermediate course — nationalized industries such as British Leyland and British Steel have been saved from collapse by taxpayers' money. Universities and other institutions of higher education have been exposed more directly to the cold winds of economic change, and like many parts of British manufacturing industry, are running well below capacity. They too will emerge transformed from the present slump. One possible and desirable outcome would be that at least some of them should be more directly involved with continuing education. If the government means what it has been saying these past several months about the importance of retraining and about its respect (or at least lack of enmity) for higher education, and because the falling inflation rate suggests that the time is fast approaching when reflationary policies will again be necessary, it will be absurd if this opportunity is let slip.

How to make dreams untrue

Although the hunt for the philosopher's stone has long since been abandoned, the hunt for its antidote, the material that turns everything into lead, continues apace, most successfully in the dealings between the British government and the nationalized industries for which it is responsible. This is the chief lesson to be drawn from the report of the House of Commons Select Committee on Energy (see page 621) on the new British nuclear energy programme. The committee plainly found its inquiry into the rights an wrongs of the proposal and infuriating experience. The British government chose not to pass on to the committee the advice provided by the Central Policy Review Staffs on the wisdom of building two reactors of the advanced gas-cooled type, thus confirming the widespread understanding that the advice was hostile to the project — and the belief that when British governments extol the benefits of open government, they are thinking of governments other than themselves. The Department of Energy, ostensibly in charge of the substantial commitment of resources entailed by the nuclear programme, is convicted of incompetence by its evidence in the House of Commons. The Central Electricity Generating Board, the nationalized electricity utility which is the constructor and operator of nuclear plants south of the Scottish border, is accused not of excusable incompetence but of unforgivable deviousness. When all the dust has settled, the chief casualty of the committee's forthright report will be the British nuclear programme, fanned to life only in 1979 from the embers of a decade's neglect.

Even after due allowance is made for the way in which select committees these days have to shout to be heard above the reports that flood from their competitors, the language used by the Select Committee on Energy will surprise many of those at whom the complaints are levelled. For the committee's chairman and deputy chairman are both members of the now defunct Select Committee on Science and Technology which, over the years, has been one of the most consistent cheerleaders for nuclear power in Britain. What has gone wrong, the luckless targets of the report will be asking? The answer is that they have compromised a good cause by arrogance, fecklessness (which is a synonym for conscious laziness) and a failure to understand what they are for. In the short run, only they may suffer. Further ahead, taxpayers (and electricity consumers) will also be the losers.

The problem of nuclear power in Britain is by now a problem in national mythology. Ever since the Tizard mission to Washington in 1941, the popular belief in Britain has been that nuclear fission was invented ("discovered" is too passive a word) in the land of Newton. The Ottawa Agreement in 1944 merely codified what most people knew, for which reason it followed as night does day that there should be a truly Britishnuclear cxplosion (in 1948) and, now, a nuclear deterrent (called Trident) whose design may be American but whose construction is beyond question British. The commercial exploitation of nuclear power in the past quarter of a century has been a cross between a fairy story and a nightmare. From time to time, radical innovations of technology have been announced, have been widely advertised to an unsuspecting press and then have been allowed to run into the sand because nobody cared about them in the sense of knowing that not merely his reputation but his pension (index-linked) would depend on the realization of his promises to the public. To everybody's surprise, however, the nuclear reactors built in the first heady days of the nuclear enterprise (with natural uranium as fuel and graphite as a moderator) are mostly still working well and helping to reduce the price paid by electricity consumers.

The select committee's forgivable irritation has plainly led it to complain most vociferously at those who sought unsuccessfully to mislead it — the Central Electricity Generating Board is the worst offender. The issue, the committee says, is whether the board can possibly be right in pretending that the nuclear power stations it plans to order in the coming decade will be economical devices for generating electricity when there is so much evidence that the board's assumptions are either untested or untestable. All kinds of questions arise. Can the board be right in guessing that the cost of producing coal (the obvious alternative to uranium) will increase by between 2.5 and 3 per cent a year in the foreseeable future? Can it be believed in quoting capital costs for building the simplest structures when it has so mismanaged the oil-fired power station still painfully half complete on the south bank of the Thames Estuary? Is it, indeed, possible to consider seriously the figures produced by the board and others, purporting to quote future electricity generating costs, if none of the interested parties turns a hair when it turns out that the estimates are wrong, and that the consumers of electricity must pay the difference? For all its rhetoric, the committee would have had a more telling case if it had been able to specify the premises on which utilities (such as the Central Electricity Generating Board) might set out to calculate the future cost of electricity. The truth is, of course, that in the real world there are none that carry conviction. What happens if there is, in the end, a war in the Middle East? It would be too late, then, to call the committee back into session.

With luck, such a drastic course will not be necessary. The committee has done what it needs to do for the decade ahead. It has thrown a spanner in works that were not working. The recommendation that the United Kingdom Atomic Energy Authority should be the chief agent of technological change in the nuclear industry, concerned principally with fast reactors and with fusion, is to be welcomed. The plea that the Department of Energy should somehow equip itself to understand the policies with which it pretends to grapple is right, but will quickly be forgotten with the connivance of the department. The crux of the issue, with which even this loudspoken committee has not come to grips, is that there is no mechanism in the present arrangement for the sponsorship of nuclear or any other kind of power that will allow that the consumer of the electricity should be king. The present British government has widely advertised its wish to reduce the public sector's borrowing requirement by inducing private investors to share some of the risk. Is it not time that it sold the Central Electricity Generating Board to somebody with a long purse — or better still, a conscience? The committee's irritation has led it correctly to pillory the board. It should go on to define criteria by which all such boards should measure their duty.

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More secrecy on cryptography research

US academics lean towards self-restraint

Washington

Mathematicians and computer scientists in the United States will soon be asked voluntarily to submit papers on cryptography and related research to the National Security Agency before publication, to see if the agency feels they contain anything that should be kept secret.

This precedent-setting system of self-censorship is being proposed by a study group set up last year by the American Council on Education to look at the growing conflict between national security and academic freedom in cryptography research.

The proposal is something of a compromise between the Defense Department's demands for strict legislation to control the publication of research results with potential security implications, and critics who argue that there should be no restriction on the publication of non-classified research.

However, several scientists warned last week that such voluntary self-regulation could lead eventually to demands for a similar approach to research ranging from lasers to integrated circuits.

Tensions between the Defense Department's security agency and sectors of the research community have grown steadily over the past few years. They stem partly from a desire by the agency to limit the spread of knowledge about virtually unbreakable codes — and the insistence of mathematicians that since many difficult mathematical problems can provide the basis for such codes, any restrictions would have a "chilling" effect on research.

Vice-Admiral Bobby Inman, director of the National Security Agency, has argued fiercely that the free publication of research results could inhibit the agency's data-gathering capabilities. Scientists reply that the codes also have important civilian applications — such as the protection of computer data — that justify their wide dissemination.

Last year these tensions rose to the surface when a computer scientist at Massachusetts Institute of Technology, Dr Leonard Adelman, found a grant application to the National Science Foundation had been passed to the National Security Agency. The agency subsequently offered to support part of his research — but on terms which would have given it the right to determine how much should be published.

The incident caused considerable embarrassment to the National Science Foundation — which protested that it had been seeking the views of the security agency on cryptography research applications for several years. It also pointed out that potential conflicts were being studied by the study group of the American Council on Education, set up at the suggestion of the National Security Agency to discuss ways of controlling the distribution of research results acceptable to the scientific community.

After a year's study, the group agreed at a meeting in Washington last week to propose a system leaving responsibility in the hands of scientists and journal editors by setting up a voluntary review system by the security agency.

According to the group's proposals, soon to be circulated in the scientific community, either a scientist or an editor could submit a paper to the agency for comments on whether it contains information considered to be a threat to national security.

If the agency had no objection, the scientist would be free to publish. If it did object, then the scientist could decide not to publish, proceed with publication against National Security Agency advice — or refer the matter to an independent, five-person committee. This would have two individuals named by the security agency, and three picked by the President's science adviser from a list submitted by the National Academy of Sciences.

In principle, officials of the American Council on Education hope that voluntary self-regulation — which would be introduced for a trial period — would avoid the difficulties of new legislation (which could run into constitutional problems over freedom of expression) while meeting the security agency's main concerns.

In practice, getting the system to work will not be easy. The first step will be for the security agency to prepare a guide to the type of research projects it would expect to evaluate. If the list is too broad, agency officials admit they could end up stifling research unnecessarily; yet if it is too narrow, they fear both that they might tip off others about their principal interests, and miss potentially valuable research findings.

There is also likely to be considerable resistance from the scientific community. Only one of the study group's nine members voted against the proposal for self-censorship; this was Dr George Davida of Georgia Institute of Technology, who found a patent application intercepted by the National Security Agency three years ago, and subsequently received a letter threatening consequences if he discussed his research with his colleagues.

Several academics, however, are worried that self-regulation would create a new category of secret research, pointing out that classified research is now banned on many campuses following the anti-war demonstrations of the 1960s.

The study group's proposals are therefore likely to generate considerable heat. But the political tide is now running in its favour and those who protest at the encroachment of security agencies on individual liberties have fewer friends in Congress than in the past. Vice-Admiral Inman has been nominated as deputy director of the Central Intelligence Agency — and remains committed to the desirability of strong government controls over potentially sensitive research.

David Dickson

Committee douches nuclear energy

The British government's 1979 statement on nuclear power, like its predecessors, is a muddle. This is the opinion of the House of Commons Select Committee on Energy, published this week. The committee asks that decisions to build nuclear plants in the 1980s and 1990s should be decided on their merits and not as part of a planned programme.

On economic grounds, the committee is sceptical about the government's programme to build 15 GW of new nuclear plant by 1992. The Central Electricity Generating Board (CEGB) comes in for particularly sharp criticism. The report cites several instances where the board's evidence on costs was misleading. It criticizes the board for basing future costs on early Magnox plants without acknowledging the effects of subsequent inflation on future capital investment, and for comparing the costs of electricity generated by different types of plant by using "highly uncertain variables" such as the average load factor of plant and future fuel and fuel cycle costs.

Most damning is the complaint that the CEGB presented international cost comparisons suggesting that a pressurized water reactor (PWR) would cost 34 per cent more to build in Britain than elsewhere. The committee says that the generating board's estimate of PWR costs are "too perfunctory" and that it is too tolerant of inefficiencies in the British construction industry. Planning permission for the first PWR plant is still to be sought. Subsequent plants will be either PWR or AGR (advanced gas-cooled reactor) depending on cost and performance.

The committee also suggests that the size of the British programme could be cut if CEGB and the South of Scotland Electricity Board reduced their planning margins for the excess capacity needed for plant failure in particularly severe winters. These have crept up to 28 and 73 per cent respectively from about 17 per cent in the

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fifteen years ago.

The electricity generating boards are not the only organizations to be criticized in the report, however. Criticisms of the Nuclear Installations Inspectorate, the independent body responsible for nuclear safety, made in Parliament last year, is reiterated. The inspectorate needs to put more effort into assessing the PWR, a design new to Britain, says the committee. Two aspects of PWR design, the integrity of the pressure vessel and problems of two phase flow in the water coolant, call for highly specialized inspectors which the inspectorate lacks. The committee recommends that the inspectorate takes on an ultrasonics expert for testing pressure vessels and that the government remedies, by means of legislation if necessary, the inspectorate's difficulty in attracting suitably qualified staff because of uncompetitive salaries.

The UK Atomic Energy Authority (UKAEA) is criticized for past bad advice to government. The committee recommends that its role as adviser to government on nuclear policy be given to the Chief Scientist's office at the Department of Energy. The UKAEA, it says, should confine itself to research on future nuclear options such as the fast breeder reactor and fusion.

One example, in the committee's opinion, was the recommendation not to consider the Canadian CANDU reactor as a possible option. The committee clearly believes that the CANDU reactor offers certain advantages over both the AGR and the PWR but realizes that the government is too committed to its present plan for an about-turn now. Instead, it asks for a study of CANDU before a final commitment to the PWR is made. **Judy Redfearn**

European Community

Research compared

Brussels

The European Community seems to have taken fright at the gap between Europe and the United States and Japan in spending on research and development. In his first speech to the European Parliament last week, the president of the European Commission, Gaston Thorn, said that in 1981 the Commission will give priority to research and development that will improve the Community's competitiveness with Japan and the United States. The latest assessment of research spending shows that in Europe as a whole, research and development expenditure accounts for 1.9 per cent of Gross Domestic Product, compared with 2.3 per cent in the United States and 2.0 per cent in Japan. If defence research is excluded, the figures are 1.7 per cent, 1.7 per cent and 2.0 per cent, indicating Japan's present dominance by this yardstick.

What, asked Vincent Ansquer, a French member of the European Parliament, can the Commission do? The answer seems to be that the Commission will hope to increase its own research spending in the next few years from 1.6 per cent to 2.0 per cent of the combined research and development budgets of the member states.

Constitutionally, the Commission is less able to influence national expenditure directly but is working on a series of studies which are to be summarized as guidelines for a common research and development policy due to be presented at the Council of Ministers in June this year.

Community research and development expenditure is assessed each year in a report by the Scientific and Technical Research Committee (CREST). Comparisons between member states have been quite influential in the past; for example, they prompted the French government to make substantial increases in its research budget in both 1979 and 1981. The latest report, still being finalized, will say that between 1979 and 1980, member governments' expenditure on research grew in real terms by at most 0.4 per cent.

During the whole of the 1970s, it is now clear, growth rates were highest in West Germany, Ireland and the Netherlands, with France, Italy and the United Kingdom below average. In 1980, Italy emerged at the top of the growth table, with an increase of 20 per cent in real terms of its research and development budget.

The CREST report uses information about the objectives of government expenditure in 1970–79 to infer changing priorities. Throughout the Community, government support for "the general promotion of knowledge", still the largest item, is declining. In stark contrast to government declarations, however, European governments seem to have allowed research contributing to industrial productivity and technological development to fall proportionally.

Increased proportions of national budgets have been spent on the exploration of the Earth and the atmosphere, the planning of the human environment and the protection and improvement of human health. The widely acknowledged need to reduce European dependence on imported energy seems not to have received its due — over the 1970s, real expenditure increased by a mere 0.4 per cent and actually decreased by 1 per cent immediately after the increase of oil prices in 1973–74. Energy research, nevertheless, has an important role in Italy and Germany, while in Denmark and the Netherlands most government expenditure continues to be "on the general promotion of knowledge".

The influence of defence research expenditure on the pattern of research in individual countries is catalogued in the CREST report. The disparity between the United Kingdom and its partners stands out. Thus British expenditure on defence research grew from 41 per cent of the government's budget in 1970 to 53.3 per cent in 1979. Only France comes anywhere near this proportion, with 35 per cent of the government's budget going to defence research in 1979. In the same year, Germany spent 11.7 per cent on defence research and the other six member countries little or nothing. The report does point out, however, that defence research brings industrial spin-off.

The outlines of the committee's picture remain clear enough. Research spending by Community governments is now increasing after the trough in 1978 but still falls short of American expenditure. (The Japanese statistics are incomplete.) In money terms, research and development expenditure in the United States in 1979 was about 1.3 times greater than the corresponding figure for the nine members of the Community. The good news, though, is that in 1980 (according to forecasts of US federal agencies) American government expenditure may have declined while that in Europe remained constant. **Jasper Becker**

Toxic chemicals

UK regulations

The long awaited and potentially contentious draft regulations by which the British government will require the notification of new chemicals were published by the Health and Safety Commission on Wednesday (18 February). The provisional timetable for discussion allows until July for comment, in which case the regulation could become law before the deadline of 18 September laid down by the European Commission. But Brussels, in this as in other matters, is running late and may not be in a position to make its 1979 directive binding on all member states until next year.

The draft regulations are substantially more stringent than the proposals described in the commission's discussion document published in 1977. The principle that they should apply only to new substances remains, but outline notification will now be required for substances manufactured or used in quantites of less than 1 tonne. Chemical manufacturers will now also be required to provide information about biodegradability, thus meeting one of the criticisms of the earlier proposals by environmentalists. One measure of the increased stringency of the draft regulations is that the estimated cost of testing a new chemical is now given as £45,000.

The proposed regulations for the United Kingdom are more stringent than those required by the European Community in that they apply to chemical intermediates as well as to products supplied to others. This is one of the points on which British chemical manufacturers are likely to concentrate in the coming months, but there will also be complaints that the proposed regulations allow the Health and Safety Executive to pass on to other European authorities information about

0028-0836/81/080622-02$01.00
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chemicals newly introduced by British manufacturers.

Full notification (from which chemicals supplied to research laboratories will ordinarily be exempt) is elaborate. Manufacturers will be required by means of an acute toxicity test to classify new substances as toxic (LD_{50} less than 200 mg per kg), "very toxic" (LD_{50} less than 25 per kg) or neither, using oral doses in laboratory rats. Ordinarily, the chemical identity of the material will have to be defined, as will the purity of the material and "the nature and the concentration of the main impurities", which may be harder.

For most manufacturers, biological tests will be the most time-consuming. Apart from acute toxicity data (with oral doses in rats, subcutaneous doses in rats or rabbits and inhalation doses in rats), notification requires a 28-day sub-acute toxicity test, acute toxicity tests with fish and daphnia, tests for skin and eye irritation and of skin sensitization and a mutagenicity test. The regulations empower the Health and Safety Executive to require, on the basis of a preliminary examination of the first set of data (which has to be completed within 45 days) further information including the results of tests for teratogenicity and carcinogenicity.

Exceptions to the strict requirement of notification include the supply of chemicals required for further development and materials supplied in quantities amounting to less than 1 tonne (where chemical identity will usually be sufficient). One possibly attractive let-out is the provision that users of such small quantities may be able to meet the requirements by submitted a sample in a container labelled "CAUTION — substance not yet fully tested".

Industrial innovation

States chip in

Washington

Support by the states for technological innovation is becoming fashionable. Echoing a theme launched in Congress and by the federal government three years ago, several states are now beginning to put political force behind legislation intended to stimulate technological innovation in local industry. Many realize that their economic future may depend on their ability to attract and keep high technology companies in competition with rival bidders. Measures are therefore being introduced at the state level comparable with the federal initiatives proposed by President Carter two years ago for stimulating investment in high technology.

Often the new measures include the provision of substantial research funds for state universities to promote research in areas of particular commercial value such as the technology of microcomputers. Many states are also trying to draw their universities closer to the research and training needs of the private sector. Last week, for example, the state assembly of New York passed a package of four bills to promote the growth of high technology industry in the state and to forge closer links between the state government, private industry and local universities.

One of the New York bills would revivify the state's Science and Technology Foundation, originally established by Governor Nelson Rockefeller in 1973 to improve the quality of university research in the state. The new bill, likely to be accepted by the state senate and passed into law, would require the foundation, now virtually defunct, to help small companies obtain federal research grants such as those offered by the National Science Foundation. The bill also provides new incentives for colleges and universities to carry out potentially commercial research, and two members with management experience in high technology companies would be added to the foundation's board. A second bill, concerned specifically with research workers at the State University of New York, would increase the proportion of profits from the successful commercial development of research results that might be kept by a faculty member.

The growing interest of all states in the development of industrial policy has prompted a synposium organized this Saturday (21 February) in Washington by the governor of California, Edmund G. (Gerry) Brown immediately before the annual meeting of the National Governors' Association. Mr Brown has recently proposed a $22 million "reindustrialization" programme for California, claiming the need to forge a new partnership between government and industry. This demand was the basis of his unsuccessful bid for the Democratic presidential nomination last year — and is expected to emerge again in the 1984 election.

Central to Mr Brown's programme is a proposal to establish a Microelectronics Innovation and Computer Research Operation. This would be based at the Berkeley campus of the University of California and would focus on basic research in microcomputers considered too costly to be carried out by individual companies. The state would allocate $7.6 million to the project, $2.6 million to be spent by the university and the rest to be matched by grants from private companies, most of which have welcomed the proposal.

Similar plans are being worked up by other states. For example, the Minnesota state legislature is considering a request for substantially increased funds from the Microelectronics and Information Sciences Center at the state university. In North Carolina, whose governor, James D. Hunt, has promised to make the state the centre of the East Coast microelectronics industry, the government agreed last summer to allocate $1.8 million to a microelectronics centre in Research Triangle Park which has already tempted General Electric to commit $50 million to an integrated circuit plant nearby.

The Californian initiative is in part defensive. Because of high real estate prices and shortages of technical staff, many states are now sending delegations to Silicon Valley to persuade established semiconductor companies of the virtues of relocating elsewhere. Ohio has already been offering attractive inducements to companies prepared to move their operations, and while Mr Brown will be talking in Washington on Saturday, a delegation from New Orleans will be in California describing the advantages of the Deep South. **David Dickson**

Telecommunications

Monopoly intact

Halfway through the committee stage in the House of Commons, the British government has agreed to amend its telecommunications bill in ways that will please British Telecom but are unlikely to reassure telecommunications equipment manufacturers. The amendment will give British Telecom, the new telecommunications authority, the right to be consulted during licensing of equipment for attachment to the public telephone network. Manufacturers, already dismayed at the modest erosion of the public monopoly promised by the original version of the bill, now fear that their freedom to develop new communications technology will be further constrained.

The bill, introduced by Sir Keith Joseph, Secretary of State for Industry, last year, is the British government's watered-down equivalent of the steps taken by the US Federal Communications Commission to stimulate innovation by closer definition of the public monopoly in telephone systems. (Similar moves are under way in West Germany.) The British bill, which would split the mail-handling and telecommunications parts of the Post Office into two separate corporations, would leave British Telecom with total control of the telephone network but shade its monopoly on the supply of equipment.

The procedures in the original version of the bill for ensuring that attachments do not interfere with the operation of the network would take control from one bureaucracy, the Post Office, and give it to another, the Department of Industry. One of the amendments has it that standards will be set by the British Standards Institution in conjunction with British Telecom and the department; licences to manufacture equipment are now to be issued by the department after taking British Telecom's advice. Manufacturers complain that standards will take a long time to set and that British Telecom, potentially a manufacturer in its own right, will retain too much control. These issues are likely to be debated further before the

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committee stage ends, probably before the Easter recess.

Under the new arrangements, British Telecom will be able to manufacture and sell attachments only through independent subsidiary companies. Financing such ventures, however, will be difficult within the government's constraints on public borrowing. This is one reason why the government has amended the bill to allow British Telecom access to private finance. It may allow the corporation to work outside the public sector borrowing requirement, an issue not finally settled by the Treasury. One difficulty is that, whatever the source of capital, the government is still ultimately the guarantor.

In the next few months, equipment manufacturers will be keenly waiting to see how the removal of the monopoly is to be staged. The plan is to have a transitional period of three years during which the Department of Industry would control the market, giving British manufacturers time to plan for free competition. Much will depend on the Secretary of State for Industry, who is given wide enabling powers by the bill. Another open question is the arrangements for value added services. The bill gives the Secretary of State power to instruct British Telecom to allow private companies to lease part of the public network for the provision of services to third parties. What Sir Keith finally decides may depend on the outcome of a study by Professor Michael Beesley of the London Business School. **Judy Redfearn**

Carcinogen criteria

US retracts

Washington

Pressed by a recent Supreme Court ruling, US occupational safety officials have agreed to relax their previous opposition to including risk assessment calculations in decisions about reducing exposure to potential carcinogens. Hitherto, officials of the Department of Labor's Occupational Safety and Health Administration (OSHA) have argued that the scientific uncertainty of risk calculations — and the lack of an explicit requirement in the agency's authorizing legislation — meant that they should not be used as the basis of policy decisions. Instead, they have maintained that that once a substance has been demonstrated as a potential hazard, exposure should be lowered to the "lowest feasible level".

OSHA is being required to shift its position as a consequence of a decision by the Supreme Court last summer to strike down regulations which the agency had proposed for limiting industrial exposure to benzene (*Nature* **286**, 97; 1980). The court ruled that OSHA had failed to demonstrate that reducing benzene exposure limits from ten to one parts per million would reduce the level of "significant risk". Following this ruling, OSHA has now announced that it is revising its generic carcinogen policy, introduced last year as a mechanism for regulating any chemical suspected of causing cancer.

Under the terms of this policy, any chemical which meets one of a number of criteria — for example, which is shown to cause cancer in one experiment with laboratory animals, and also produces a positive result in a short-term test — is automatically labelled a carcinogen.

Originally, this would have been sufficient to invoke automatically the requirement that exposure be reduced to the lowest feasible level. Now OSHA officials have agreed to include consideration of whether the chemical poses a significant risk — using a variety of data to make this judgement, including court interpretations of previous rulings, OSHA's previous experience in regulating toxic substances and "prudent occupational health policy".

According to an announcement made in the *Federal Register*, three aspects of the benzene decision will be incorporated in the cancer policy. First, the significance of existing risk must be estimated before issuing a carcinogen standard; second, the exposure level must be set at the lowest feasible level which is "reasonably necessary or appropriate to eliminate significant risk"; and third, that OSHA must consider "all relevant evidence" in making such determinations.

This change in policy represents a significant shift from OSHA's previous position. The agency's former head, Professor Eula Bingham, has described risk assessment calculations as "abhorrent" to public health administrators; and the agency has strongly resisted pressure from government economists to push occupational regulations into a neatly quantifiable mould.

At the same time, the shift does not go as far as many in industry would like. Their demand is for full-scale cost-benefit analysis of all occupational health and safety regulations, based on the argument that federal controls have become a major economic burden. Although the Supreme Court, in striking down the benzene regulation, did not make the widely expected pronouncement on whether cost–benefit was required to demonstrate that a new regulation was "reasonably necessary", it is expected to do so in ruling on another case which has been brought against OSHA on cotton dust standards.

In any case, agency officials expect that a demand for full-scale cost-benefit analyses of future regulations — with the requirement that the least expensive option be adopted — will be one of the first ways in which the new Administration will try to meet its election promise of reducing the force of government regulations.

The labour movement, which has consistently argued that cost–benefit analysis is little more than a smokescreen designed to cover the relaxation of safety and health controls, is preparing for a fierce and lengthy battle. It has already complained about one of Mr Reagan's first anti-regulation acts, withdrawing new regulations published by OSHA in the last days of the Carter presidency which would have required the labelling of all hazardous chemicals used in the workplace.

David Dickson

Greenhouse effect

Act now, not later

Stockholm

The theme that the time has come for policy-makers to take account of carbon dioxide when drawing up energy policies ran through an Earthscan meeting on carbon dioxide, climate and energy last week. But speakers' conviction that action should be taken now was matched by their caution in predicting exactly what would happen if carbon dioxide emissions continue to increase.

The fundamentals are broadly agreed. The pre-Industrial Revolution atmospheric concentration of carbon dioxide was about 290 parts per million, and the burning of fossil fuel has been the largest single factor contributing to concentrations which are expected to double by the middle of the next century, assuming a 2 per cent annual growth rate in the use of fossil fuels.

The reality of the greenhouse effect was also common ground between the speakers. Predictions about specific climatic changes in specific parts of the globe were, however, more equivocal.

Current models, according to Professor Bert Bolin of the University of Stockholm, are inadequate but "they are all we have". The models are especially inadequate in dealing with the role of clouds and the interaction of the atmosphere and the oceans. Dr Tom Wigley of the University of East Anglia pointed out the difficulties of distinguishing the signal from the noise: knowing when variations in regional climates stem from a particular factor such as carbon dioxide and when they are simply part of the continual natural variation.

Professor S.K. Sinha from the Indian Agricultural Research Institute in New Delhi was the only speaker daring to be at all optimistic, and even his belief that agriculture could adapt to climate changes was conditional on fruitful research being done on water management, the identification of new genotypes more tolerant of temperatures 3–4°C greater than at present and on higher crop yields with a smaller input of fossil fuels..

The most eloquent plea for energy policies to take account of carbon dioxide came from Gus Speth, chairman of President Carter's Council on Environmental Quality. In the last days of the Carter presidency, the council urged that "full consideration" should be given to carbon dioxide in the development of

0028-0836/81/080624-02$01.00

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United States and global energy policy.

One of the first priorities in a preventive strategy would be to decide "what level of atmospheric carbon dioxide should be considered a prudent upper bound". Should we allow an atmospheric build-up of, say, 50 or 100 per cent over pre-industrial levels? The upper bound would carry with it implications for both developed and developing countries and would raise questions about the sharing of the fossil fuels whose use would be allowed.

Speth was not particularly optimistic about the chances of getting these things done. It is very hard to provoke an international response to an intangible problem whose consequences are not yet even predictable. Dr Thomas B. Johansson of the University of Lund could offer a little encouragement in that the sorts of energy policies desirable from a carbon dioxide point of view were also becoming increasingly necessary, in Sweden at least, for economic reasons. **Wendy Barnaby**

Plasma research

German setback

The Max-Planck-Institute for Plasma Physics at Garching has been forced to make a major reappraisal of its future. Because of government financial restraints (*Nature* 5 February), the institute's next major project, the "Zephyr" tokamak, has been cancelled. The institute, one of the three leading fusion laboratories in Europe, is now planning that its next project should be financed by a redistribution of resources within its total budget of about DM100 million a year.

Zephyr had been planned to leapfrog the joint European tokamak machine called JET, now being built at Culham in the United Kingdom, and would have experimented with ignited plasma. The hope now is that a redistribution of the budget will yield between DM20 million and DM100 million over the next seven years to build a less ambitious machine.

The reassessment at the institute will be carried out under new management. Last week, Professor Klaus Pinkau was appointed director of the laboratory. Although not a plasma physicist but a cosmic ray physicist, he has considerable experience of international collaboration, the politics of big science and the management of scientific institutions. He has been the director of the Max-Planck-Institute for Extraterrestrial Physics, next door at Garching, and is chairman of a committee reporting to the federal government on the merits of ten big science projects which, curiously, did not include Zephyr.

Pinkau said last week that it was dangerous to make scientific institutions too dependent on "annual changes" in the financial position of governments but, also, that budgets should not grow too fast. Certainly this year's changes at the Institute for Plasma Physics will give him pause: not only has Zephyr been deleted but the proposed budget for 1981 has been cut by 15 per cent. This trimming of sails may give the laboratory a sense of realism, persuading it that it cannot alone compete with JET, but a tough internal struggle seems inevitable between the advocates of an upgraded stellarator and a mirror machine. The edge might be taken off this battle if the new project were adopted by Euratom as a "preferred project", in which case between 10 and 20 per cent of the cost could come from Brussels. The advocates of an improved stellarator point to their success last year when the existing machine at Garching, Wendelstein VIIA, was used to show that a stellarator plasma could be held stable in conditions only previously obtained in tokamak machines. The same series of experiments created conditions of plasma density and confinement time more stringent than those reached by tokamaks of similar size, apparently putting stellarators back in business.

Tokamaks are in fact beginning to lose favour because of the various difficulties (fatigue and maintenance, for example) expected to arise in power reactors. Diversification is therefore considered prudent, whence current interest in stellarators and mirror machines. The Lawrence Livermore Laboratory in California is in fact building a mirror machine (the Mirror Fusion Test Facility) in which a large long solenoid is plugged at the ends with magnetic quadrupole mirrors. Garching cannot hope to compete with Livermore in money terms but, some argue, could attack the principles of such a device. This, broadly, is the second proposal being considered at Garching. A decision is expected in the middle of the year. **Robert Walgate**

Tokamaks and stellarators

In both tokamak and stellarator, plasma is confined to a torus by two superposed magnetic fields. One field runs around the torus, along its long circumference; the other winds round and round the small circumference of the plasma ring.

The first field is created in both tokamak and stellarator by a coil wrapped round the small circumference of the torus. The second is created differently in tokamak and stellarator.

In the tokamak, it is the result of a current carried in the plasma itself; in the stellarator it is the result of a component of current in the outer coils along the major circumference of the torus. As the plasma ring is electrically isolated, the tokamak must create the longitudinal current in the plasma by a transformer effect, and so must be pulsed.

The stellarator, on the other hand, can in principle be run statically. Early tokamaks — which were invented in the Soviet Union — were successful principally because of their large "aspect ratio" (major torus radius over minor), it is now believed, rather than because of any intrinsic merit of the tokamak design; and the need for pulsed operation is seen as a disadvantage in the construction of a true reactor, where the variation of thermal, neutron, and magnetic stresses could increase material fatigue.

Princeton perplexities

There are slippery hands and red faces at the Princeton Plasma Physics Laboratory, where a gaggle of lawyers is trying to decide who was responsible for dropping a 350-ton generator component during the construction of the Tokamak Fusion Test Reactor (TFTR).

The accident happened in December, when the outside stator of the vertical axis generator was being lowered into position. A crane bearing broke and the stator fell 15 feet, damaging both itself and the central rotor.

The incident is not expected to have a significant impact on the construction schedule for the TFTR, which will be used to achieve energy breakeven for the first time and to investigate the engineering features of large fusion systems. A second generator, already in place, will be able to supply sufficient energy for the test reactor, at least in the early phases of operation. However, a detailed study will now be necessary to determine whether the generator can be repaired — or whether a replacement is needed, which could cost up to $2 million, and take some time to deliver.

Present construction schedules anticipate that the TFTR will come into full operation in July or August 1982. This is about seven months later than the original completion date of December 1981, due largely to delivery delays on some of the major components — in particular the toroidal and ohmic field coils used to contain the plasma — which have presented more technical difficulties than expected.

Officials at Princeton say that they do not foresee any insuperable problems, as most of the technology is "state of the art". However, the delays will inevitably add to the construction costs, which are expected to exceed the predicted $284 million by about 10 per cent.

More than five subcontractors may be involved in the heated debate over the responsibility for December's accident. A report is expected shortly from the Department of Energy, which is expected to identify errors of judgement responsible for the crane overload. However, with large insurance sums at stake, any such conclusion is likely to be contested — and will almost inevitably end up in the courts.

0028-0836/81/080625-01$01.00

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CORRESPONDENCE

Wrong rodents

SIR — The discovery that several cell lines claimed to be derived from patients with Hodgkin's disease in fact come from an owl monkey [1] emphasizes the vital importance of quality control in biological research.

Several cases of genetic contamination of inbred strains of laboratory mice and rats have come to light recently, and although these are not associated with any known scientific fraud, they have led in some cases to a considerable disruption of research projects. In the United States, for example, one large commercial breeder appears to have been selling "inbred" Lewis (LEW) rats which are not histocompatible, leading to a flood of complaints from research workers and inevitably casting doubt on the validity of some published work. In Japan, a survey of over 100 colonies of inbred mice [2] has shown that about 10 per cent had been genetically contaminated either recently (with continued genetic segregation at some loci) or in the more distant past. Indeed many of the major sublines of common inbred strains such as C3H and C57BL have arisen as a result of genetic contamination in the past [3].

In the United Kingdom a voluntary genetic monitoring scheme for commercial breeders was started about five years ago by the Medical Research Council Laboratory Animals Centre and several cases of genetic contamination have been discovered since that time [4]. Both of the main UK suppliers of inbred mice and rats took these results so seriously that they have established their own in-house genetic monitoring programmes run under the direction of consultant geneticists. The International Committee on Laboratory Animal Science (ICLAS) is preparing a handbook on methods of genetic monitoring, and is considering the establishment of some international reference centres for genetic monitoring. In the meantime research workers are strongly urged not to take the authenticity of the strains that they use entirely on trust. If they purchase animals from commercial breeders they should demand to know what steps the breeder is taking to monitor the stock. Those who breed their own strains should remember that accidental contamination can occur at any time, and they should check their stock using skin grafting or some other suitable method (such as the study of biochemical [5] or immunological markers [6] or morphological features such as mandible shape [7]) as a matter of routine.

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1. Dickson, D. *Nature* **289**, 227 (1981).
2. Nomura, T. *et al. Proc. 7th ICLAS Symposium, Utrecht* (Fischer, Stuttgart, in the press).
3. Festing, M.F.W. *Inbred Strains in Biomedical Research* (Macmillan, London, 1979).
4. Festing, M.F.W. & Lovell, D. *Proc. 7th ICLAS Symposium, Utrecht*, (Fischer, Stuttgart, in the press).
5. Groen, A. *Lab. Anim.* **11**, 209-214 (1977).
6. Festing, M.F.W. & Totman, P. *Lab. Anim.* 14, 173-177 (1980).
7. Festing, M.F.W. *Nature* **238**, 351-352 (1972).

French museums

SIR — Seen from this side of the Channel, Halstead's criticisms of the new exhibition policy of the British Museum (Natural History) appear extremely unjust (*Nature* **288**, 208; 1980), especially towards the staff who contributed to such clear and remarkable presentation. In France, there is no discussion of that sort, because most of our natural history museums simply have no exhibition policy at all, and are nothing but museums of museums!

Last November, when Halstead was writing his venomous letter, I visited the two new exhibits (on dinosaurs and on the evolution of man) at the British Museum, and was amazed to see how much the public liked them, in particular the children, who seemed very receptive to the logic of cladistic analysis. Halstead's long attack against that method of tracing the interrelationships of living beings deserves more comment. He claims that cladistic analysis, which is the basis of the British Museum exhibits, does not take into consideration the concept of gradualism, according to which evolution can be presented as uninterrupted series of species, older ones being ancestral to younger ones. Hence his accusation against cladism, which is, according to him, trying to make evolution fit Marxist views on the history of societies! Halstead presents gradualism as evidence, and cladism as a crime against evidence. But where is the evidence of gradualism? There is almost none, or rather it is everywhere one wants to see it.

Cladists do not deny that a species can give birth to daughter species, but they claim that in practice, it is impossible to determine the fossil ancestor of a species, since the ancestor is devoid of the derived characteristics of the daughter-species in question. Ancestor–descendant relationship is not, for cladists, a necessary statement, and the history of a group is better expressed by sister-group relationships, often illustrated by a cladogram. Many biostratigraphers are now convinced that successions of species based on stratigraphic distribution are nothing but illusions and that, in reality, each older species is extinguished by the younger one, whose centre of origin may be far from the locality studied. So, the subversive palaeontologists who are supposed to pervade the British Museum, knife between teeth, are simply those who consider that gradualism gives an illusory precision, which is unnecessary for tracing the course of evolution of a group. If these new views are congruent with vague ideological inferences on human societies, so what?. . . After all, the history of human societies often shows a succession of leaps, be they revolutions, wars, epidemics, or changes in climate. But major changes in human societies are seldom decided by the societies themselves. The societies and their chiefs want gradual change (compare the terms "changes in continuity" or "permanent revolution" used by right-wing and left-wing politicians respectively), but in the event, circumstances provoke sudden changes.

In France, some authors are now attacking Darwin (see P. P. Grassé, *L'homme en Accusation,* Albin Michel, Paris) because he is supposed to have inspired Hitler and still inspires the deeply anti-Marxist sociobiologists. Since cladism is also directly descended from Darwin's ideas on phylogeny and systematics, it can just as well be accused of supporting extreme right-wing ideologies.

In sum, even if some cladists claim that this method of analysis is more consistent with their political convictions, it is simply ridiculous to condemn it on the basis of such spurious arguments, or because Halstead's political opinions are different. The British educational system can be proud of the British Museum's exhibitions which will certainly teach future generations of biologists and palaeontologists the principles of phylogenetic reasoning. Perhaps this is just what Halstead is afraid of?

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More museums

SIR — Halstead's well publicized reactions[1-6] to the exhibition policies of the British Museum (Natural History), to non-gradualistic hypotheses about the evolutionary process, and to his own mistaken perceptions of cladistic analysis are unfortunate[7-9]. His notion that these views somehow abet non-scientific creationist metaphysics or logically support Marxist dialectics is preposterous. Whether Popper[10], Patterson[11], or Miles (cited by Halstead in ref.4) have said that the evolutionary concept is also metaphysically based and whether this is true are beside the point. Equating Marxist political theory either with cladistic analysis or with so-called macroevolutionary hypotheses is as wrong as social Darwinism was. It needs to be driven home, as Patterson has done[9], that cladistic analysis is about pattern, not about any particular hypothesized evolutionary process, although most phylogenetic systematists do hold that descent with modification is an economical explanation of the existence of biological pattern in general. These systematists also define "related" to mean "genealogically related." In all six publications Halstead confuses cladograms with evolutionary trees. For instance, a steady theme is, "there is no place for ancestral species in a cladogram"[2]. Again, "it is axiomatic, therefore, that no species in the fossil record can be ancestral to any other nor can one species evolve directly into another"[5]. Halstead has not understood that cladograms do not assume that species are ancestors, but neither do they deny possible ancestry when a taxon lacks known derived characters. Assumptions of ancestry are appropriate for trees but are not made in cladograms.

Cladistic analysis parsimoniously estimates relatedness and is therefore testable. The British Museum (Natural History) is to be congratulated for bringing epistemology into its exhibits and teaching visitors that science is a method, not a body of revealed knowledge. That the museum may also need to discuss various hypotheses of evolutionary process[12]

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and to address other needs as well, such as the public's thirst for discussion of functional anatomy, ecology, behaviour, and so on[13], is also evident.

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1. Halstead, L.B. *Nature* **275**, 683 (1978).
2. Halstead, L.B. *Nature* **276**, 759-760 (1978).
3. Halstead, L.B., White, E.I. & MacIntyre, G.T. *Nature* **277**, 176 (1979).
4. Halstead, L.B. *New Scientist* 17 July 1980, 215-217 (1980).
5. Halstead, L.B. *Nature* **288**, 208 (1980).
6. Halstead, L.B. *Nature* **289**, 106-107 (1981).
7. Patterson, C., Forey, P.L., Greenwood, P.H., Miles, R.S. & Jefferies, R.P.S. *Nature* **277**, 175-176 (1979).
8. Fink, W.L. & Wiley, E.O. *Nature* **280**, 542 (1979).
9. Patterson, C. *Nature* **288**, 430 (1980).
10. Popper, K. *Unended Quest* (Open Court Publishing, La Salle, Illinois, 1976).
11. Patterson, C. *Evolution* (Cornell University Press, 1978).
12. Donovan, D.T. *Nature* **289**, 105-106 (1981).
13. Benton, M.J. *Nature* **289**, 106 (1981).

SIR — I have read with diligence the continuing controversy sparked by Halstead. Thus far I have had difficulty achieving a precise understanding of it. Now, however, certain matters are clear (*Nature* 1/8 January, p.106.) To sum up: Halstead dislikes the new exhibits at the British Museum (Natural History), and he would convince other persons that they, too, should dislike the exhibits. The argument he gives, as far as my evaluation goes, passes not from the ridiculous to the sublime, but emanates entirely from the low end of that spectrum. Knowing Halstead to be usually of good cheer and judgement, I am led to suspect that not all is as it might seem — that the root of his dislike is not to be found brewing in a pot pourri of punctuated equilibria, Marxism, scholastic death, etc. Rather, his dislike may stem, as seems to me, from a sense of loss of "the fossil record" — the ultimate source of the truth of evolution as rendered by a professional class of fellow-specialists. To the dismay, sometimes acute, of the more clerically minded members of this profession, cladistics treats fossils in a secular fashion — not as revelation but as some among many other biological specimens subject to interpretation that is apt, indeed expected, to be diverse, especially with respect to details (for example, the true nature of the "Petralona skull"). As reasonable as this treatment might seem to the outsider, the emotional effect within such a palaeontologist involuntarily confronted with cladistics (as I have witnessed on more occasions than I care to remember) is not unlike that apt to be experienced by a fundamentalist minister who has forced upon him uninvited the notion that the Bible is just one book among many. Suffice it to say that more than one kind of church has been built upon rock.

So what now? Here in the States creationists dislike the museums' secular exhibits on evolution and the schools' secular treatment of that subject. In Britain a palaeontologist dislikes secular exhibits on the "fossil record". The forms are similar, but the substances at first glance seem utterly different. Palaeontology, after all, is nominally a science, and a rational mind can easily defend it as such. The problem I have with Halstead's defence, if I may term it that, is reconciling it with a standard of rationality.

GARETH NELSON
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Genes and racism

SIR — Steven Rose notes in his recent letter (*Nature* 22 January, p.335) that a National Front journal *New Nation* has claimed to find support for racism in my writings on sociobiology, as well as in those of Dawkins and Maynard Smith. Rose calls on the latter two authors to dissociate themselves from such misuse, although curiously he does not extend the same invitation to me. To keep the record straight, I am happy to point out that no justification for racism is to be found in the truly scientific study of the biological basis of social behaviour. As I stated in *On Human Nature,* "I will go further and suggest that hope and pride and not despair are the ultimate legacy of genetic diversity, because we are a single species, not two or more, one great breeding system through which genes flow and mix in each generation. Because of that flux, mankind viewed over many generations shares a single human nature within which relatively minor hereditary influences recycle through ever changing patterns, between the sexes and across families and entire populations".

If there is a possible hereditary tendency to acquire xenophobia and nationalist feelings, it is a *non sequitur* to interpret such a hypothesis as an argument in favour of racist ideology. It is more reasonable to assume that a knowledge of such a hereditary basis can lead to the circumvention of destructive behaviour such as racism, just as a knowledge of the hereditary basis of haemoglobin chemistry and insulin production can lead to the amelioration of their pathological variants.

I now call on Professor Rose to consider these and similar arguments raised in my writings. It is my hope that he will not confine himself, as he has in the past, to arguments that link sociobiology to racism and thus to continue to abet the very misuse which he piously claims to deplore.

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Origin of cancer

SIR —John Cairns' article on "The origin of human cancer" (*Nature*, 29 January 1981, pages 353-357) dismisses the importance of chemical mutagens in human cancer aetiology on, we believe, very tenuous grounds. He argues that there must be a single underlying mechanism for tumour production and that this is not through point mutation. As your leading article (*Nature*, 5 February 1981) quite rightly points out, why should there be a single mechanism for cancer development? Indeed, can one say that cancer is even a single disease? Cairns' article appears to us to be a simplistic approach to a complex problem. We all would like to have the answer to how a cancer cell develops, but can one say that Cairns' article will lead us any nearer to the truth? Why should workers in the field of chemical carcinogenesis abandon a large body of work which may be getting somewhere near to establishing why populations differ in cancer incidence, and substitute a hypothesis of genetic transposition bearing in mind that, as Cairns writes, it is not yet clear whether transposition is important in vertebrate development. Should we abandon the somatic mutation theory of cancer for a hypothesis with no experimental evidence? Furthermore, why should mutagens/carcinogens only act through point mutations? It is well recognized, as Cairns states, that agents reacting with mammalian cell DNA can cause gross effects such as chromosome aberrations, sister chromatid exchanges, deletions, and so on. Just because one measures DNA reaction in bacteria with a point mutation system does not mean *a priori* that this is the mechanism of action of these chemicals in mammalian cells. After all, bacteria don't even get cancer. Bacteria are merely used for carcinogen screening because they are cheap and mutations are easy to score.

Where Cairns' article is mischievous is in suggesting that people are wasting their time looking for carcinogens in the environment and then, having identified them, seeking to reduce exposure. The fact is that if people would stop exposing themselves and their immediate families to mutagens/carcinogens in the form of cigarette smoke, a large proportion of cancers would be prevented (besides a large proportion of coronary heart disease, lung disorders, etc.). If Cairns accepts that smoking is bad for one, why should he not accept that other environmental insults might also be carcinogenic? To say that animal carcinogens induce mainly liver cancer, that humans don't normally get cancer of this organ, and therefore animal liver carcinogens have nothing to do with human cancer shows a complete unawareness of chemical carcinogenesis. Most chemicals which have been identified as human carcinogens do *not* give the same spectrum of tumours in animals as in humans, for example benzidine causes bladder cancer in man but liver cancer in animals.

Cairns' main argument for DNA reaction being unimportant in carcinogenesis is the finding that in xeroderma pigmentosum patients, few if any internal cancers have been seen. Why should one expect an increased incidence of lung cancer, for example, in these patients? Is the skin the same as the lung in its function, biochemistry, enzyme profile, etc.? Why should we expect the mechanism of skin cancer to be the same as for internal organs? Do xeroderma pigmentosum patients live in the same environment as the normal population? I don't think one has to indulge in "special pleading" to support the case of somatic mutation as being important in cancer production. The majority of facts available tend to support the somatic mutation hypothesis. When there are sufficient data available to overthrow this hypothesis, then is the time for it to be abandoned. In the meantime, those of us working in chemical carcinogenesis will carry on identifying carcinogens in the environment and recommending that exposure be reduced. Whether society (or Cairns) listens to us is entirely up to them.

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The Great Badger Debate

What follows is a brief summary of the report Badgers, Cattle and Tuberculosis, *published by the British Ministry of Agriculture, Fisheries and Food last October and a comment by its author, Lord Zuckerman, on various criticisms that have been raised, some of them in* Nature

Report and implications

Bovine tuberculosis has been recognized as a serious problem in the United Kingdom (and also a source of human tuberculous infection) at least since 1913, when steps were first taken to control the disease. Farmers were encouraged to register herds free from tuberculosis by means of a premium price for the milk they produced, and infected cattle were also slaughtered compulsorily. But bovine infection continued, with unexplained "breakdowns" even in attested herds.

The link between bovine tuberculosis and badgers was first recognized in 1971, with the discovery of tuberculous lesions in a badger found dead. The organism responsible was found to be *Mycobacterium bovis*, that responsible for bovine tuberculosis. It now appears to be common ground that badgers can be infected by this organism. In 1975, ministry officials began controlling badgers found within a specified radius of infected herds of cattle by the use of cyanide gas. These procedures were abandoned in October 1979, but have been resumed in South West England since the publication of the Zuckerman report.

The transmission of tuberculosis from badgers to cattle is accepted in the report, which says that viable *M. bovis* have been recovered from grass sprayed with the urine of infected badgers (but more easily in the winter than the summer). The report also says that during the period in which control by gassing was carried out, "185 out of 2,432 badgers that had been collected in the vicinity of 477 farms where breakdowns had occurred" turned out to be infected with *M. bovis*. The report also records how, in New Zealand, the opossum was discovered to be a reservoir of bovine tuberculosis, and gives (in its Appendix III) accounts of how, at several farms in South West England, infection in badgers has been linked with breakdown among herds of cattle.

The South West of England sustains a third of the United Kingdom's cattle, and also dense populations of badgers. It is common ground between Lord Zuckerman and those who have criticized his report that badgers are also prevalent in parts of England, where bovine tuberculosis is rare. Lord Zuckerman concludes, in his report, that the population density of badgers determines both their susceptibility to infection and the risk of transmission to cattle. The frequency of infection among badgers in the South West of England has been found to be as great as one in five in some places.

Among the recommendations in the report were the resumption of badger control by gassing in the South West, the investigation of the incidence of tuberculosis among badgers in contiguous parts of the country, financial support for field studies of the British badger population, the publication of annual reports of the progress of the control campaign and a more thorough review of what will by then have been learned of the badger problem after an interval of three years.

Zuckerman's case

The 6 November 1980 issue of *Nature* (p.6) carried a brief account of a report on tuberculosis (TB) in badgers and cattle which I had prepared for the Minister of Agriculture, Fisheries and Food. The report was written in response to a request for an assessment of the soundness or otherwise of the scientific evidence on which the ministry based the view that badgers in parts of the South West of England constitute a reservoir of the bovine tubercle bacillus. Because of its statutory responsibility to suppress TB in cattle, I was also asked whether the government was justified in its policy of destroying badgers where there was presumptive evidence of the presence of tuberculous badgers that might be responsible for outbreaks of TB in neighbouring cattle.

During my investigation I had consulted several scientists engaged in the study of animal disease, as well as practising veterinary surgeons. All agreed with the findings of the ministry's scientific and veterinary officers with many of whom I had conferred. My own review of the evidence left me satisfied that badgers in certain areas of the South West are heavily infected with the bovine tubercle bacillus, and sometimes transmit the disease to cattle grazing on pasture that has been contaminated with their sputum, pus, urine and faeces. I uncovered not a vestige of scientific foundation for widely publicized statements from "protestors" to the effect that none of the story was "true" and that, in effect, the whole thing was a hoax perpetrated by government servants.

It was, of course, unlikely that a fresh display of the facts would necessarily succeed in "converting" the opposition. But it came as a surprise to read first, in an unsigned article in *New Scientist* (4 December 1980, p.619), and then in a letter published in *Nature* intended as a corporate statement by The Mammal Society (11 December 1980, p.532), that the conclusions of my report, in which I had assembled not only more, but also more up-to-date, information on the subject than had been made available before, were questionable on scientific grounds. Apart from agreeing that "the badger is a major reservoir of bovine TB in certain limited areas of South West England, and hence a potential danger to the cattle in those areas", the society asserts that my report gave a "biased interpretation of the evidence", that "many of (my) conclusions were not justified" by the data which I had presented, and that "factually misleading statements" therefore needed to be corrected.

The report

My main conclusions, set out in the report, were first, that "by any rational epidemiological standards, badgers (in some parts of South West England) now constitute a significant reservoir of the bovine strain of the tubercle bacillus". The Mammal Society thinks otherwise. It declares that "there is no scientific evidence to justify" the view that badgers in the affected areas constitute "a highly infected population". If the society is sufficiently well informed to make this extraordinary claim, the onus of providing proof, as Dr Plowright has already pointed out (*Nature* 1/8 January 1981, p.8), is on them.

Next, the Mammal Society was, to say the least, irresponsible in their quotation of what was in the report. I did not "overlook" the fact that during the first seven months of the moratorium the percentage of badgers with TB had fallen slightly in Cornwall at the same time as it had increased in Gloucestershire and Avon; this is precisely what my figures revealed. Nor did I interpret these or any of the other available figures in the "black or white" way that the Mammal Society suggests I did and, now in reverse, that it itself does. The interpretations I provided were the best that could be put on the figures, and were carefully qualified. Conclusions contrary to those I drew from the facts would have been absurd.

The Mammal Society is not criticizing when it merely repeats what I emphasized: namely, that the disease in badgers occurs only in "pockets". But it is falsifying what I did write when it says that I had asserted that badgers in the affected areas of the South West are spreading TB to badgers in other areas. What I wrote was that no one knows how one pocket of infection in badgers relates to another (paragraph 134 of the report). And I did not say that TB *is* (my italics) a major hazard to the survival

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of the badger (even if it clearly is to the badgers in the areas already affected). What I said was, were the disease "to take hold in badgers in other parts of the country" "there is no saying what the consequences would be . . . as they relate to the survival of the badger".

In considering the persistence or extinction of a disease in a given locality, the statistical laws governing its spread must be borne in mind. If the average number, x, of new infections from a unit which becomes infected is less than 1, the disease will die out. If greater than 1, the disease, once established, will spread. However, if x is not markedly greater than 1, there is still a good chance that a single infection will not result in its establishment; but if repeated new infections occur, then eventually one of them will be successful in establishing the disease.

With badgers two separate units are involved: first, the individual within a badger sett, and second, the setts themselves. As Dr Plowright has re-emphasized, the epidemiological conditions are highly favourable for transmission of TB within a sett. Transmission between setts seems to depend very greatly on such factors as density of population, contiguity of setts, and fighting between members of different setts. In an area which adjoins another where the disease is endemic, there will be a continuous flow of infection from the infected area to the border setts of the other area. But if x for the second area is less than 1, the infection will not spread through the whole area.

Infectivity

It is obvious that the period during which a unit remains infective depends on the nature of the disease. For a disease which kills quickly or in which there is complete recovery without the unit becoming a carrier, the period will be short. In a disease such as TB it will be considerably longer. Note also that the value of x will change with time, both because of the direct effect of the disease on population density and of acquired immunity, and also because of other unrelated factors.

I have discussed these matters with Dr Frank Yates of Rothamsted. In his view, my supposition that bovine TB may have been prevalent in badgers throughout the country before its virtual eradication from cattle is almost certainly justified (paragraph 133 of my report). After its eradication from cattle and the consequent absence of cross-infection the disease died out in areas of the country in which x was less than 1, as it seems to have done in most of the UK.

Dr Yates has also pointed out that once the disease has died out in an area, this area will remain free as long as no new infection is introduced, but if owing to an increase in the density of population or for other reasons x becomes greater than 1, the disease, once established, will then spread until the whole area is infected. The question put by the Mammal Society as to why areas in other parts of the United Kingdom with a badger population density which is presumed to be as great as in the South West were not infected, seems basically simple. Either x has remained less than 1, or they have just been lucky.

Before turning to certain further information about the prevalence of TB in badgers that has become available since my report was written, it is necessary to refer to what I said about the incidence of breakdowns in cattle, since this is also something about which the Mammal Society is critical.

The campaign to suppress bovine TB resulted in a decline in the incidence of reactors in cattle herds, not only in the South West, but also in the rest of England. This the Mammal Society acknowledges. But, as Dr Plowright has already pointed out the society failed to note in its statement the critical fact that while the incidence of herd breakdowns has fallen dramatically in all parts of the country once the culling of tuberculous cattle was made compulsory in 1950, in the affected areas of the South West it has consistently remained more than five times the national average. This difference was the essential reason for the search for a "cause", and for the resultant finding of a reservoir of the bovine tubercle bacillus in badgers in the affected areas. And here is where the Mammal Society's criticisms have gone wildly astray. Their suggestion that one possible interpretation of the figures given in the Report was that unknown "subtle factors" resulted in a decline of TB in both cattle and badgers throughout the country, and that gassing had little significant effect on the overall timing or rate of decline is, as Dr Yates has shown (*Nature* 22 January 1981, p.218), based on a very superficial and distorted use of the evidence. His more detached presentation clearly indicates that the gassing campaign appears to have had a considerable effect in reducing the very high incidence of herd breakdowns in the worst affected parts of the South West. What the Mammal Society did was lump together in a graph my figures for TB in badgers in all the affected counties of the South West, whereas as Dr Yates has shown, there is a radical difference when one compares the parallelism in the prevalence of TB in badgers and the incidence of breakdown in cattle as between Cornwall and Gloucestershire on the one hand, and Devon, Dorset, Somerset and the rest of England on the other.

In its haste to criticize, the Mammal Society has also clearly failed to realize that the data assembled in my report were the best available at the time, and that more would be published as they became available — I have recommended annual reviews. The society's charge that it was "manifestly untrue" that herd breakdowns reflected a high local prevalance of TB in badgers was based on the

Lord Zuckerman (right) with Agriculture Minister Peter Walker and the report

figures for Cornwall, where only 15 per cent (51/340) of the breakdowns were attributed to badgers. What the society failed to bring to the attention of its readers — their examination of the evidence must indeed have been cursory if they themselves failed to notice it — is that, as Dr Yates did note, in Cornwall 69 per cent (234/340) of the breakdowns were classed as unknown, whereas in the rest of the South West (plus Sussex) only 26 per cent (107/417) were so classed, 67 per cent (280/417) being attributed to badgers.

Figures

Before rushing into print the society might indeed have enquired further about the Cornish figures. I have now obtained a breakdown by years which gives:

Cornwall	*Unknown*	*Badgers*
1972–73	110	–
1974–75	88	10
1976–78	36	41
	234	51

The explanation is simple. Badger investigations were only begun in Cornwall as a routine measure in 1974. The progressive reduction in the proportion of "unknowns" indicates their increasing thoroughness. I am also informed that for the whole of Cornwall 72 per cent of the 51 outbreaks attributed to badgers were within 2 miles of setts containing infected badgers, and 33 per cent were within half a mile. For the West Penwith area the corresponding percentages were 97 and 47.

The data which I had when I wrote my report related to only the first seven months of the period of the moratorium (Table 18 of my report). I have now been provided with figures for the first twelve months during which no new gassing operations were carried out. During the year, 611 badgers were autopsied in connection with official investigations to determine the source of herd breakdowns in the affected areas of the South West, and 73 (12 per cent)were found to be infected. Needless to say, there were differences between the counties concerned, but since the significance of these differences could only be determined through a detailed analysis aimed at revealing their relation to specific

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herd breakdowns, they hardly merit discussion.

More important is the information that is now available about the herd breakdowns that occurred in the affected areas in the first twelve months of the ban, about which nothing positive could be said on the basis of the data that were made available to me when I drew up my report, and in which reactors were found in only 0.7 per cent of the 17,411 herds that were tested. Between April and September a further 6,845 herds have been examined, bringing the total up to 24,256. The percentage of reactor herds had risen substantially in comparison with the first seven months of the moratorium in every one of the affected counties of the South West. Overall, the percentage for the period April to September was 2.2, double that for the period January to September 1979. For the year of the moratorium as a whole it was some ten times higher than for the rest of England.

The anonymous article in *New Scientist* (4 December 1980) referred not only to the Mammal Society's statement, but also (1) to certain critical comments by Dr Hans Kruuk that follow the same line as the society's statement; and (2) to an assertion by Miss Eunice Overend, described as an "enthusiast" of badgers, about the speed with which TB develops in badgers; she is quoted as saying that while TB may develop quickly in captive badgers, "it wouldn't happen in the wild". In answer to an enquiry I put to her, she also challenges the views of veterinarians about the speed with which the disease can take hold in cattle.

I have no doubts about Miss Overend's sincerity, but her "facts" happen to be wrong. Advanced TB lesions have been found in the lungs of wild badger cubs ranging in age from four to nine months, which had clearly been infected by routes other than bite wounds, and it is estimated that at least some of these cubs would have succumbed to the disease in a matter of weeks.

Anomalies

Of course, there are still many "anomalies" in the story, which further enquiry might elucidate. But every one of the so-called anomalies referred to in the Mammal Society's manifesto, all its speculative questions, "why this, why not that", are, as Dr Plowright has pointed out, mentioned in my report. Setting them out as a form of criticism adds nothing to the story. I hope that the further enquiries which I suggested should be undertaken to clear up a number of "unknowns", and which the Minister of Agriculture, Fisheries and Food, in his statement to Parliament said would be started as soon as the necessary arrangements can be made, will be entrusted to experienced and competent hands.

So much for the "scientific" criticisms that have been levelled at my report. No new facts have been brought forward. Nothing that has been said would make it anything but irresponsible for the government to devise a policy of action for the suppression of bovine TB, to which it is statutorily committed, other than what it is doing. Unfortunately, the Mammal Society's statement has been used as part authority for a press-release by the UK branch of the World Wildlife Fund, in which the government's acceptance of my report is in effect condemed. This was issued without any consultation with the Animal Health division of the Ministry of Agriculture, Fisheries and Food (MAFF).

As a scientist I have been a dedicated supporter of the conservation movement since before the Second World War,and as a scientific adviser to government I played a part in organizing our own formal institutions for the protection of the environment. I therefore find it regrettable that the only way I could have found favour with those who are concerned to obstruct the government's anti-bovine TB policy would have been either to suppress, or to fudge, the evidence which had been gathered by those scientists primarily concerned over many years with the problem of TB, not only in cattle, but also in badgers (as well as in those luckier representatives of the wild fauna of the country which, by comparison, are relatively resistant to the infection).

Mammal Society

The Mammal Society was one of the bodies which submitted written evidence to help my enquiry, citing Stephen Harris and Dr Kruuk as its two authorities since, so the Society said, they were "the two principal non-MAFF scientific researchers on badgers in this country". Presumably both helped in the drafting of the corporate statement which was designed to correct my "factually misleading statements". Since there can be few land-mammals that have generated as much proprietary interest as has the badger among the handful of naturalists who have added to our knowledge of the ways of the animal, I should note that I cannot find in the zoological literature any paper by Harris on badgers. Dr Kruuk's present studies relate to the animal's natural history, and I referred (paragraph 141 of my report) to a proposal which he had already published, that "pastures in infected areas should be so dressed as to reduce drastically the population of earthworms which they sustain", commenting, however, that I did not know "whether there is a way of doing this without detriment to the cattle which also forage the pastures that would be treated", nor "what those concerned with the welfare of birds — which also eat worms — would think about the measure which Dr Kruuk has proposed" Before turning to the badger, Dr Kruuk studied the behaviour and ecology of gulls and carnivores — mainly hyenas. For this work he was awarded the Zoological Society's Scientific Medal in 1975.

The errors in the Mammal Society's statement suggest that its badger panel did not include experienced epidemiologists or veterinary scientists. That it should have done is clear. For contrary to what has been said to me by the society's president, Dr Ernest Neal, the study of mammals is not just "a rather narrow field of interest" that can be covered by watching the behaviour of animals in the wild. Whether or not it was intended, the misleading impression which the statement has made on the media, and on those who now clamour against the government's decision, was that the Mammal Society is a body that can make pronouncements on animal disease which, if anything, might have been expected from the Royal College of Veterinary Surgeons, or from a commitee set up *ad hoc* by the Royal Society.

Of course members of the Mammal Society are free if they so wish to criticize the scientific validity of the data that I collected and to question my interpretations, but by insisting that it has the right to make a corporate, but anonymous, statement of the kind it has the young Mammal Society has, in my view, done itself harm. Here it is useful to refer to the main editorial in *Nature* of 11 December 1980 (p.525) entitled "Code of conduct for national academies", which pointed out that even "The Royal Society has no means by which it can reach a corporate opinion on any substantial issue of public policy . . . for example, say, government policy on nuclear power stations or whether manufacturers of drugs should in future be liable for side effects". Of course, the Royal Society could set up committees to look into such matters, but then its members would be named, and those Fellows of the Society not on the committees would certainly not be committed to whatever recommendations were put forward. If it wishes its published statement to be considered further then the Mammal Society should clearly name the "experts" who were responsible for its drafting. Dr Yates suggests that what the Mammal Society has done illustrates "the way in which distorted and partial graphic presentation can be used for propaganda purposes". It might have been more charitable to say that what was done was done out of ignorance. But then, as Gunnar Myrdal has put it, one has to remember that ignorance, like knowledge, can be steered for a purpose.

Not only was there no scientific value in the Mammal Society's statement; it was clearly the work of only a handful of its members, none of whom, so far as anyone can tell, had ever worked on the epidemiology of tuberculosis in animals. It is unfortunate that the society's action should have encouraged those who protest against MAFF's findings and policies in the belief that they have scientific support for the new wave of opposition which they have already mounted.

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NEWS AND VIEWS

Too soon for the rehabilitation of Lamarck

CAN acquired characteristics be inherited? The arresting letter from Gorczynski and Steele (on page 678 of this issue) says that there are circumstances in which they can be. The interest of the observations is beyond dispute. The days have long since gone when dark lamarckian issues such as these could be raised respectably. Even in primary schools, or at least in those not yet captured by the creationists, young people are now told that the giraffe did not come by its long neck because earlier generations of giraffes had stretched their necks in an attempt to eat the leaves of ever-taller trees, and had been able to pass on the characteristics thus acquired. So how can it be that dark lamarckian evidence can be uncovered at this late stage in the study of inheritance? That is what many people will be asking. The simple answer is that it is too soon to know what to make of the observations now reported. On such an important issue, no single set of observations can be sufficient to overturn the accepted view of how inheritance is organized in living things. Independent confirmation is now the most urgent need. So much has been clear since the first article on this subject by Gorczynski and Steele a year ago (see R.B. Taylor *Nature* **286**, 837; 1980). By all accounts, confirmatory evidence has been hard to come by. Some reports of failures to confirm seem destined to appear in the next few months. Gorczynski and Steele, like their would-be critics, have chosen an intricate field in which to work. For the time being, however, the results they report must be put firmly in limbo. They do not constitute a proof that Lamarck has been right all this past century and a half, but only a question to be pondered.

The lamarckian heterodoxy, it must be remembered, had respectable beginnings. Lamarck had the wit to recognize the need for an explanation both of the diversity of species and the evidence for evolutionary processes uncovered (by the Lyells of this world) from the sedimentary rocks. At the outset, the notion that characteristics once acquired might be inherited cannot have failed to be seductive. What could have seemed more sensible, in the first half of the nineteenth century, than that a giraffe with the good sense to stretch its neck in the search for better food might produce offspring with similar characteristics? The difficulty, from the beginning of the Lamarckian thesis, is that it has never amounted to an explanation. If the giraffe came by its long neck thanks to the diligence of its ancestors, why does not a woman whose arm is amputated produce one-armed children? Lamarckians, both ancient and modern, have not been able to answer such questions.

Historically, none of this has mattered very much. Darwin's explanation of both diversity and the appearance of change, or evolution, turned out to be a better explanation. Both diversity and change, said Darwin, could be explained by the mechanism of evolution by natural selection. Longer-necked giraffes might be fitter in the sense of being the more able to propagate themselves. So much is now accepted, yet darwinian scholars are forever gleefully discovering in *The Origin of Species* or other darwinian texts evidence of Lamarckism in Darwin himself. So what? Formally, Lamarckism and Darwinism are not mutually exclusive. One does not disprove the other. Although Darwin showed that natural selection is the predominant process in evolution, the data available in the second half of the nineteenth century could not have excluded the admixture of a modicum of lamarckian inheritance. The contemporary orthodoxy stems not from Darwin but from the turn of the century. The rediscovery of mendelism provided a mechanism by which variations could arise and thus be selected. Weissman's doctorine that there is a sharp distinction between the cells of the germ line and the soma meanwhile outlawed Lamarck. In modern language, according to Weissman, the genotype has a decisive influence on the phenotype, but the phenotype can affect the genotype only because the phenotypically fitter individuals are more adept at the propagation of their own kind. So far, everything that has been learned about the mechanisms of ineritance in animals and plants is consistent with both Darwin and Weissman.

Laboratory experiments in which viruses have been shown to carry new information into the germ line are nevertheless a tantalizing hint that there may be exceptions to the rule. Gorczynski and Steele challenge Weissman's doctrine, not the belief that darwinian selection is predominant in the evolution of species. Their latest experiments are an extension of those described a year ago. Then, they claimed that male mice made tolerant to a single histocompatibility antigen produced, from normal mice of the same strain, offspring that were also tolerant of the same antigen, at least in the first and second generations. The acquired characteristic (tolerance of a specific antigen) appears to be inherited. The latest series of experiments begins with male mice made tolerant of two antigens. Again, the data seem to suggest, tolerance to both antigens appears in the first and second generations. On the evidence available — and the numbers are only small — the two acquired characteristics are transmitted independently to the first and second generations of mice. Gorczynski and Steele put forward a possible explanation of their results. Weissman's doctrine has been violated, they say, because some naturally occurring RNA virus may have picked up the altered genes from the immunological system of the mice made tolerant to histocompatibility genes, and may have transferred these to the germ line. The results, they say, "demand an explanation not offered by conventional neo-darwinian genetics".

Not everybody will agree. For one thing, the experiments so far described are incomplete. If, indeed, a somatic mutation has been transferred from the soma to the germ line, then it should be possible to select both male and female mice of the first generation carrying the gene for tolerance and demonstrate that, thereafter, the mice breed true. In reality, the frequency of tolerant individuals is less in the second generation than the first, suggesting that whatever is being inherited cannot be fixed in the germ line. Moreover, there are other possible explanations for the results so far reported. One suggested by R.B. Taylor is that the mice may have inherited not the gene for tolerance but a gene for the production of the antigen used for the induction of tolerance. Other explanations will emerge. While so little is known of the mechanism of tolerance in these mice, and while conventional explanations have not been ruled out, it is premature to call for a recasting of genetics, "neo-darwinian" or otherwise. And the experiments need to be repeated.

Gorczynski and Steele will no doubt

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regard this response to their interesting article as toffee-nosed. Others, sadly, will go further and say that even a single case of the inheritance of an acquired characteristic implies that contemporary genetics is a house of cards. That is an outrageous leap ahead.

Even if the data now reported are confirmed, the explanation of them offered by Gorczynski and Steele may turn out to be no more than an exception that proves the conventional rule. For if it requires the extreme procedures that have been used in these experiments to provoke such an indecisive example of lamarckian inheritance, may not the generality of Weissman's doctrine be strengthened? Even that speculation is however premature. It will be time enough to worry about the rehabilitation of Lamarck when and if the data now reported are independently confirmed. □

Gamma-ray astronomy

from R. D. Wills

THE spectral range of gamma-ray astronomy is very broad. At a recent meeting* results were presented and discussed which span seven decades of photon energy from about 10^5 to more than 10^{11} eV. The techniques described differ considerably along the energy range, but essentially two approaches are adopted to overcome the problem of atmospheric absorption. At the lower energies, where the fluxes are relatively higher, instrumentation is carried above the atmosphere by balloons or satellites. Above about 10 GeV it is advantageous to measure at sea level or mountain altitude the secondary particles generated by interactions of the primary radiation in the upper atmosphere. These secondaries can be detected over a wide area enabling very low fluxes to be measured.

Most of our knowledge about high-energy (30 MeV to 5 GeV) gamma rays has been obtained from the NASA satellite SAS-2 (C. E. Fichtel *et al. Astrophys. J.* **198**, 163; 1975) and its ESA successor COS-B (L. Scarsi *et al. Proc. 12th ESLAB Symp.* ESA *SP-124*, 3; 1977). The latter has made the first complete detailed survey of the Milky Way in high-energy gamma rays (H. A. Mayer-Hasselwander *et al. Ann. N. Y. Acad. Sci.* **336**, 211; 1980). On behalf of the Caravane Collaboration, H. A. Mayer-Hasselwander (Max-Planck-Institut für Extraterrestrische Physik, Garching bei München) reported that, up to the end of 1978, 83,233 gamma rays had been recorded from the galactic disk (latitudes $|b| < 12°$) and outlined the current analysis of their spatial distribution and energy spectrum. A. W. Strong and A. W. Wolfendale (University of Durham) reviewed the results at intermediate latitudes ($10° < |b| < 20°$). As in the lower-latitude range, the gamma-ray intensity is well correlated with the various tracers of the constituents of interstellar matter. The use of galaxy counts as a tracer of the 'total gas' enabled the gamma-ray emissivity of the interstellar medium to be obtained. Its energy dependence showed clearly the importance of the contribution of cosmic-ray electrons to the generation of gamma rays. G. F. Bignami (Istituto di Fisica Cosmica del CNR, Milan) drew attention to low-latitude correlations with galactic structures on a variety of scales, and pointed out that inconsistencies between the gamma-ray map and Gould's Belt could be explained in terms of the newly discovered 'Dolidze Belt' (M. V. Dolidze *Sov. astr. Lett.* **6**, 51; 1980).

Several speakers discussed the localized gamma-ray sources, of which 25 are listed in the 'Second COS-B Catalog' (B. N. Swanenburg *et al. Astrophys. J.* in the press). P. A. Caraveo (Istituto di Fisica Cosmica del CNR) described a search for X-ray counterparts in the error boxes of some of the unidentified sources, whose positions are known only within a radius of about 1 degree. On the basis of their narrow latitude distribution R. Buccheri (University of Palermo) proposed that at least a significant part of them could (like the only two unambiguously identified sources, PSR 0531 + 21 and PSR 0833–45) be very young pulsars, but ones not yet known to radioastronomers. A complementary suggestion by T. Montmerle (Centre d'Etudes Nucléaires de Saclay) was that a class of gamma-ray sources is extended sources, associated with selected active regions in the Galaxy, like the ρ Oph cloud or the Carina complex.

Extragalactic gamma rays were reviewed by D. Ramsden and A. J. Dean (University of Southampton). In the high-energy range only the quasar 3C273 has been detected (G. F. Bignmai *et al. Astr. Astrophys.* in the press), though upper limits exist for the fluxes from many active galaxies and some galaxies in the local group (G. F. Bignami *et al. Astrophys. J.* **232**, 649; 1979; A. M. T. Pollock *et al. Astr. Astrophys.* in the press). The Seyfert galaxy NGC 4151 has been detected below 20 MeV in balloon experiments (F. Perotti *et al. Nature* **282**, 484; 1979) but lack of confirmation from other experiments indicates that it may be variable at these energies, as it is in other parts of the electromagnetic spectrum. In this energy range it is possible to detect gamma-ray line emission due to nuclear interactions or antiparticle annihilation (R. Ramaty and R. E. Lingenfelter *Nature* **278**, 127; 1979). The experimental status of such measurements was reviewed by L. E. Peterson (University of California, San Diego), while a theoretical overview was given by R. Ramaty (NASA/Goddard Space Flight Center). Both speakers mentioned the detection of spectral lines in transient (burst) events. Those of 19 November 1978 and 5 May 1979 exhibited a feature at 0.4 MeV which is believed to be gravitationally redshifted positron-annihilation radiation, probably from the vicinity of a neutron star. G. Vedrenne (Centre d'Etude Spatiale des Rayonnements, Toulouse) described how the accuracy of locating bursts by triangulation had been improved (down to 1 arc min), using the long baselines available from interplanetary spacecraft such as Helios, Pioneer Venus, Venera and ISEE-3. However, no firm conclusion on the nature of burst sources could be drawn from the distribution of their locations, nor from the burst size ($\log N - \log S$) distribution.

Gamma rays of energy greater than 10^{11} eV can be detected at ground level by virtue of the Cherenkov light from the small air showers they generate in the Earth's atmosphere. K. E. Turver (University of Durham) and T. C. Weekes (Smithsonian Institution, Mount Hopkins Observatory) described recent developments in this field and emphasized the possibility of extending the technique to lower energies using multi-element detector arrays. Of the sources already detected, new results on Cygnus X-3 were presented by A. A. Stepanian (Crimean Astrophysical Observatory) and N. A. Porter (University College, Dublin). Reviewing investigations of pulsars, B. V. Sreekantan (Tata Institute for Fundamental Research, Bombay) remarked especially on the results from Ootacamund, where pulsed emission from PSR 0531 + 21 had been detected in February 1977, but despite increased sensitivity, not in the succeeding two winters (B. N. Bhat *et al. Astr. Astrophys.* **81**, L3; 1980). There was some speculation as to whether this variability might be related to a change in the 50–3,000-MeV light curve which COS-B had detected between 1975 and 1979 (presented on behalf of the Caravane Collaboration by R. D. Wills, Space Science Department of ESA).

Future prospects for gamma-ray astronomy were reviewed by C. E. Fichtel (NASA/Goddard Space Flight Center). In the next decade satellites such as the Soviet/French 'Gamma-1' and NASA's

*The Royal Society's meeting for discussion on 'Gamma-Ray Astronomy', arranged by the British National Committee on Space Research, was held in London on 27–28 November 1980. The proceedings will be published in the *Philosophical Transactions of the Royal Society*.

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0028-0836/81/080632-02$01.00

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Gamma-Ray Observatory should carry experiments which will have over an order of magnitude greater sensitivity than those flown so far while in some parts of the energy range substantially improved energy and angular resolution will be achieved. Such experiments should greatly enhance our knowledge of several astrophysical phenomena especially those associated with compact objects, astrophysical nucleosynthesis, galactic structure, the origin of cosmic rays, active galaxies and universal matter–antimatter symmetry. Meanwhile the meeting learned from R. D. Wills (Space Science Department of ESA) that COS-B was still recording good data after more than 5 years in orbit and that ESA's Science Programme Committee had recommended the continuation of its mission into 1981. □

Diffusionless reactions and crystal engineering

from J. M. Thomas

DIFFUSION, which is the spontaneous kinetic drift of atoms, ions and molecules, is a phenomenon so inextricably mingled with a wide range of chemical phenomena that it is difficult to imagine that there could be a class of chemical transformation in circumstances where even the idea of diffusion is superfluous. The migration of ions in solution or in solids, the dissolution of an electrode in an electrolytic cell, the transport of reactants to, and products from the surface of a heterogeneous catalyst are all well known examples of chemical processes in which diffusion plays a dominant role. Indeed there are many kinetic situations — the recombination of iodine atoms in the gas phase is a renowned example — where the diffusion of the reactants is rate-limiting. Under which set of circumstances, therefore, is diffusion of such little consequence?

It is now clear that very many highly efficient and selective chemical reactions may proceed smoothly over a time scale of minutes or hours *in the solid state* without the need for prior diffusive motion. So frequently has it been remarked that chemical reactions are sluggish in solids because diffusion in the solid state is itself a very slow process, that it often comes as a surprise to discover that crystals of certain organic molecules can be induced to react rapidly, and in a preordained fashion, even at cryogenic extremes. The secret is to design these organic molecular crystals — to 'engineer' them — in such a way as to pack together potentially reactive monomer units which can subsequently be 'compelled' to undergo preferred reactions, which entail no diffusion but merely minor reorganization — a slight rotation or a subtle shift of part or all of the reactant molecules. Such reactions, which are termed topochemical, yield clean products, the chemical nature and crystallinity of which are governed more by the molecular environment within the parent crystal than by the intrinsic reactivity of the molecule.

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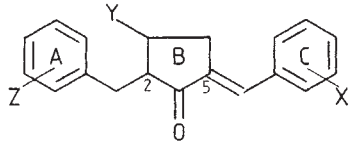
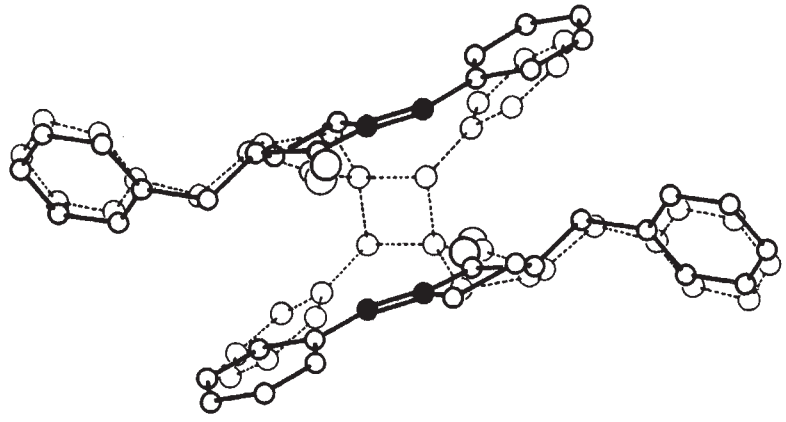
Crystal engineering consists essentially of selecting molecules of a given class and functionality according to their size, shape and stereochemistry. They are then 'persuaded' to crystallize into certain desired crystallographic space groups; and within those crystals, by appropriate stimulation — UV irradiation, for example — diffusionless, intermolecular reactions are allowed to proceed.

Many aspects of this new approach to synthetic chemistry are an echo of the past. Kohlschutter (*Z. anorg. allg. Chem.* **105**, 121; 1918), the first to use the term topochemistry, realized that certain host crystals, notably graphite, dictate the degree of topotaxy associated with a solid-state reaction. Topotaxy refers to the mutual relationship between the unit cell axes of the daughter and parent crystals — in this case the graphite intercalates (for example, graphite–ferric chloride or potassium graphite) and the graphite, respectively. The late Gerhardt Schmidt, who was amongst the first to talk of crystal engineering, along with his associates at the Weizmann Institute, made many noteworthy contributions to this field (see Schmidt *Pure appl. Chem.* **27**, 647; 1971; Cohen and Green *Chem. in Britain* **9**, 490; 1973; and Thomas *Phil. Trans. R. Soc.* **A277**, 251; 1974). They and others have shown how various tactical substitutions in a molecular framework can be used as a steering device for preferred crystallization of a given desired crystal structure. The same approach has proved successful in persuading certain diolefins to crystallize with photoreactive rather than photostable structures, an approach which, in turn, has led to the solid-state synthesis of optically active molecules and many other novel products (Nakanishi *et al. Proc. R. Soc.* **A369**, 307; 1980).

Recently the delicate interdependence of crystal structure and reactivity in a family of related molecules derived from the skeletal framework shown below



has been traced (Jones, Nakanishi, Theocharis and Thomas *J. chem. Soc. Chem. Commun.* 610; 1980). Various substitutions for X, Y and Z in the rings C, B and A were made and the photoreactivity of the monomer ascertained. The nature of the molecular packing was determined from the X-ray structure of each monomer crystal. It transpires that when the product (dimer) molecule occupies a similar volume within a crystal, and has a closely similar shape, as an adjacent pair of monomer molecules (an incipient dimer), single-crystal → single-crystal chemical transformation (without phase separation) can take place. In the figure below the adjacent molecules 1 and 2 (see framework with X, Y, Z all = H; this compound is 2-benzyl-5-benzylidene-cyclopentanone), which together constitute an incipient dimer, can undergo very minor movements at their peripheries during the course of a



Two neighbouring molecules of monomer (in which bonds are drawn with full lines) are so disposed with respect to one another in the 'engineered' crystal that they readily link up when irradiated with UV light to form a dimer. The four-membered cyclobutane ring (dotted bonds) is formed from the pairs of carbon atoms (shaded) linked originally by ethylenic bonds. Note how little the peripheral atoms (the benzene rings) move during this diffusionless reaction.

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photodimerization that converts the doubly bonded carbon atoms (shaded) into a cyclobutane ring. In this crystal, monomer molecules pack so closely to one another — the ethylenic plane-to-plane distance is only 3.80 Å — that dimerization proceeds freely to completion within the crystal matrix over a period of several hours.

Such changes are of considerable theoretical interest in that they permit, in principle, the atomic rearrangements to be monitored directly (at the diffusionless extreme) during the course of chemical reaction (Nakanishi, Jones, Thomas, Hursthouse and Motevalli; in the press). But such transformations have a wider relevance. New avenues of solid-state synthesis are opened up, by capitalizing on the idea that a potentially labile guest, when incarcerated within the well defined cell created by the crystal — be it of the pure material or of a host environment into which the guest has been buried on crystallization — has few options open to it. Through the agency of an externally applied stimulus, such as UV irradiation, the guest can to good advantage be converted into a more 'agreeable' state and then liberated (by dissolution for example) for appropriate utilization.

The sequential acts of molecular imprisonment and release, when prisoner and prison are carefully matched, can clearly be used to considerable advantage. Recent reviews (see Thomas *Pure appl. Chem.* **51**, 1065; 1979; Green, Lahav and Rabinovich *Acc. chem. Res.* **12**, 91; 1979) display the extent to which progress has been made in this direction. □

Habitat and the spread of rabies

from P.J. Bacon and D.W. Macdonald

An international workshop on rabies, habitat classification and fox populations was held at Oxford recently*. As the behaviour of vectors of rabies, most notably the red fox, *Vulpes vulpes,* is known to be affected by the habitat and as the epizootiology of rabies appears to be affected by landscapes on both a fine and a gross level, it was hoped that a meeting of zoologists, epidemiologists and geographers could spark off new approaches to understanding and combatting a disease which has so far thwarted most attempts at control.

The first problem to be tackled was the classification of habitats. A robust method of describing landscapes is necessary not only for understanding the way foxes and rabies behave in differing habitats, but also for extrapolating from small study areas to the continental scale at which disease control must work. R.G.H. Bunce (Institute of Terrestrial Ecology, UK) described a classification method that was intended, in part, to enable the findings of detailed ecological studies at specific sites to be generalised to the whole of Britain. A wide variety of ecological parameters, such as soil profiles, natural vegetation and agricultural crops, determined by field-survey, are highly correlated to topographic features that can be measured from Ordnance Survey maps: these correlations can then be used to predict the factors from map measurements of areas not surveyed. The method accurately predicts, for example, acreages of various crops and potential primary productivity and an attempt is being made to produce a map of potential (that is, 'carrying-capacity') fox densities for the whole of Britain (Macdonald, D.W., Bunce, R.G.H. & Bacon, P.J. *J. Biogeography* in the press). The predictions are supported by the known fox densities in some areas although they cannot yet incorporate local variations in human hunting pressure. P.J.A. Howard, (Institute of Terrestrial Ecology, UK) reviewed methods of classification (that is categories concerned with natural divisions within the data) that were appropriate for habitat description; and concluded that while a suitable stratification into some type of category was desirable to permit efficient sampling of habitats, the aim of prediction might be better served by statistical methods using regression of the original data on variates important to the aspects being predicted. Thus, for example, it might be better to compare fox densities to measures of the landscape (for example, percentage of woodland, percentage of cereal crop) rather than to ordinal categories into which the landscape had been classified (for example, 'woodland', 'arable land' and so on). His recommendations received support from U. Emanuelsson (University of Lund) predictions of bird breeding densities in Swedish Lapland using formulae derived from regression of known densities against measures of landscape, such as area of woodland, altitude, and so on.

The aspirations of the workshop were well illustrated by quantitive investigations of the relations between landscapes and rabies spread. Using data on case-occurence made available by L. Andral (Centre d'etudes sur la Rage, France), F. Ball (University of Oxford), looked for relations between the velocity of the front wave of rabies to a set of data (collected by himself and P.J. Bacon) describing the habitats in which those cases occurred. Ball posed three questions (a) how does the behaviour of the front wave of rabies epizootic vary with habitat, (b) is infection better described by nearest neighbour models, that is with one resident fox passing the disease to its territorial neighbours and its own group members, or by dispersing fox models, that is, those taking into account the movements of many kilometres made by young male foxes, and (c) would a knowledge of habitat effects increase one's ability to predict the likely spread of the disease? He demonstrated that variations in front wave velocity can be linked to measures of the landscape, which correlate with fox densities. Examples of important measures are altitude (there are few foxes on mountain tops), railway lines (which follow fertile valleys and hence perhaps represent ribbons of high fox density, while also channeling fox movements) and vineyards (which are associated with broken hilly terrain suitable for foxes — indeed, biblical foxes often ate grapes and present day ones certainly eat blackberries). Map-data can thus be used to explain a significant amount of the historical variation of front-wave velocity in France, and similar measurements in areas in advance of the wave-front might have predictive value.

J. Ross (Imperial College London), applied B. Sayer's (Imperial College London), approach to the French data supplied by L. Andral and demonstrated the effects of rivers and motorways in diverting the path of the epizootic. Neither are readily crossed by foxes so movements tend to be along, not across them. From the case incidence data she drew trajectories of the disease front as it spreads across France month by month. When these trajectories are superimposed upon a geological map it is evident that the disease has followed a belt of limestone running south west across France. It seemed plausible that the rich soil of this limestone country was mirrored, through high primary productivity, in high fox populations. She also showed that rabies spread through areas of different landclasses (as described by Ball) had different temporal characteristics. These differences seemed largely associated with the presence or absence of upland pastures — habitat which might be good mouse-hunting country for foxes.

*The *Workshop in Rabies Spread, Habitat, Classification, and Fox Populations* was held at Keble College, Oxford from 15 to 19 September 1980. The sponsors were the Nuffield Foundation and the Insititute of Terrestrial Ecology. A full report of the meeting (with references and a list of participants) will be edited by the organisers and will be obtainable from The Librarian, Institute of Terrestrial Ecology, Merlewood Research Station, Grange over Sands, Cumbria LA11 6JU.

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0028-0836/81/080634-02$01.00

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Considerable debate revolved around the extent to which sophisticated analyses could be extended, considering the uncertainties of rabies surveillance. The reporting of cases is much more efficient along the epizootic front than in enzootic districts where familiarity soon produce negligence. Rather dismaying disclosures about the consistency of the data on which statisticians must work were endorsed in a devastating analysis by A. Wandeler (University of Berne) of the biases in the hunting figures frequently employed as indirect indicators of fox numbers. Factors such as the phase of the moon affect hunters' behaviour, and hence the bag records! It was widely agreed that every effort should be made to standardise the recording of rabies incidence, even if only in sample districts.

Students of fox ecology were not the only biologists present, since the principles underlying analyses of habitat utilisation are widely applicable. H. Kruuk, (Institute of Terrestrial Ecology, UK), showed how the dispersion of habitat patches (and hence available food in the form of earthworms) determined the size of badger *(Meles meles)* territories and their local population density. Earthworms can only be captured on warm and humid nights when they are tempted onto the surface. From night to night, with shifting wind direction for example, different fields will temporarily become bountiful foraging sites. Kruuk argued that badger territories must contain sufficient foraging sites so that at least one will yield worms irrespective of changes in the wind. Territory size, and ultimately population density, is thus a function of the dispersion of potential earthworm patches — a factor that can to some extent be predicted from maps. When the meeting learnt from Wanderler (University of Berne) that badgers had been more involved than previously thought in rabies spread in Switzerland it was realised that these findings were of practical as well as theoretical importance. Kruuk's findings in Scotland could be compared with those of I. Lindsay and D. Macdonald (University of Oxford). They had examined the habitat components of badger territories in Buckinghamshire and found, for example, that territory size varied with the acreages of cereal land they encompassed. These arable crops are unimportant in the badgers' diet and can be interpreted as 'dead space' in the badgers' territories. S. Harris (University of Bristol), found urban-dwelling badgers living at extraordinarily high densities. Indeed, the participants collectively blanched at the prospect of rabies control in urban areas, because it was apparent that the densities of both wild and domestic vectors are extremely high and almost unassailable by conventional control methods. The highest fox densities recorded were from suburbs where Macdonald has found home ranges as small as 10 hectares. In the same habitat foxes were living in social groups — another factor which doubtless would effect rabies epizootiology.

Two schools of thought emerged on the most useful way to proceed — one was that fox population dynamics were so complicated that effort might most profitably be directed to seeking broad relationships between general features of the landscape and rabies spread, while the opposing view was that the data on rabies spread were so coarse and inconsistent that direct and detailed study of foxes was more likely to yield a better understanding of the factors affecting the pattern of rabies spread. In either case, concensus was reached that the most crippling obstacle was our ignorance of the behaviour of rabid foxes and the extent to which it differs from that of healthy animals. □

100 years ago

The Singular Methods of Travel the Wagtail adopts to Cross the Mediterranean Sea.

One evening we were sitting at the foot of the pyramid of Cheops, sipping our cup of fragrant Mocha and in jolly conversation, rolling up clouds of blue smoke from our Korani cigarettes. We were waiting for the sinking of the sun to make our return to Cairo. The deep silence of the surrounding desert possessed something uncommonly solemn, only now and then disturbed by the cry of the hoarse fishhawks far above us. Still higher the pelicans were grandly circling. Their flight, though heavy when seen from anear, possess a majesty in the distance attained by no other bird. Right before us several wagtails were hopping around and 'tilting'. They were quite tame, and flew restlessly hither and thither. On this occasion I remarked, 'I could not quite understand how these birds could make the long passage of the Mediterranean.' Sheik Ibrahim heard this from our interpreter. The old Bedouin turned to me with a mixture of French and Arabic as follows, which the interpreter aided us to fully comprehend:-

" 'Do you not know, Hadretch (noble sir), that these small birds are borne over the sea by the larger ones?'

"I laughed, as did our friends; for at first we thought we had misunderstood him; but no: the old man continued quite naturally:-

" 'Every child among us knows that. These little birds are much too weak to make the long sea journey with their own strength. This they know very well, and therefore wait for the storks and cranes and other large birds, and settle themselves upon their backs. In this way they allow themselves to be borne over the sea. The large birds submit to it willingly; for they like their little guests, who by their merry twitterings help to kill the time on the long voyage.'

"It appeared incredible to us. We called to a pair of brown Bedouin boys, pointed out the wagtails to them, and inquired:-

" 'Do you know whence come these small birds?'

" 'Certainly,' they answered. 'The Abu Saad (the stork) carried them over the sea.'

"At supper, in the Hôtel du Nil, I related the curious story to all present, but naturally enough found only unbelieving ears.

"The only one who did not laugh was the Privy Councillor Heuglin, the famous African traveller, and, excepting Brehm, the most celebrated ornithologist of our time for the birds of Africa. I turned to him after the meal, and inquired of his faith. The good royal councillor smiled in his caustic way, and with a merry twinkle remarked: 'Let the others laugh: they know nothing about it. I do not laugh, for the thing is known to me. I should have recently made mention of it in my work if I had had any strong personal proof to justify it. We must be much more careful in such things than a mere story-teller or novel-writer; we must have a proof for everything. I consider the case probable, but as yet cannot give any warrant for it.'

"My discovery, if I may so call it, I had kept to myself, even after Heuglin had thus expressed himself, and would even now maintain silence on the subject had I not recently discovered a new authority for it."

I read lately in the second edition of Petermann's great book of travels the following:-

"Prof. Roth of Munich related to me in Jerusalem that the well-known Swedish traveller, Hedenborg, made the following interesting observation on the Island of Rhodes, where he stopped. In the autumn tide, when the storks come in flocks over the sea to discover them. Once he followed a flock of storks, and as they lighted he saw small birds fly up from their backs, which in this manner had been borne over the sea. The distance prevented him from observing to which species of singing birds they belonged."

We regret that the Lords should have thrown out the Bill on Tuesday for the Opening of Museums and similar places on Sundays. As the *Times* very well puts it:— "The gravity of the question is that London has in its midst people to whom anything of the nature of intellectual toil — and prolonged sight-seeing is of that character — is essentially irksome. But they are human beings, and not lost to all salutary influence. It would be folly to despair of making the Sunday more tolerable than it is to them. Our climate does not often admit of men and women sitting out of doors talking or listening to elevating music. Some substitute must be found to put on on equality with the people of more sunny lands. It is the task of true friends of the working classes to suggest means by which, without any revolution in national ideas as to the sacredness of Sunday, they may be enabled to taste those simple and primitive pleasures — for example, the pleasure of pure repose of mind and body, or that of hearing music — which all, even the untutored, can enjoy.

From *Nature* **23**, 24 February, 387, 1880.

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The expanding science of photobiology

from John Jagger

Photobiology is concerned with the vast range of actions of nonionizing radiations on biological systems — from photolysis of simple molecules in pure solution to the effects of sunlight on human skin and problems of world food production through photosynthesis. The common thread that held the five hundred participants at the recent International Photobiology Congress at the Louis Pasteur University, Strasbourg, together is an interest in the fundamental biophysical mechanisms involved in the interactions of light and living matter. In line with this orientation is the increasing use of sophisticated physical techniques, such as picosecond dye-laser pulse studies of electron energy exchange between chlorophyll pigments in chlorophyll–protein complexes (J. Breton and G. Paillotin, Centre d'Etudes Nucléaires de Saclay; N.E. Geacintov, New York University), and picosecond kinetic measurements at liquid-helium temperature of proton translocation in the retinal molecule (M. Chabre, Grenoble).

Reversible photochemical reactions are often used by biological systems to sense changes in the environment. The most obvious and certainly the most sophisticated such system involves the protein rhodopsin pigments used in **animal vision**. Photoisomerization of the retinal prosthetic group of rhodopsin leads to an alteration of the receptor cell membrane potential, but the intermediate steps are not understood. Since the cell membrane is not connected to the disks that contain the rhodopsin, there must be a diffusible transmitter, and this is now believed to be cyclic GMP, rather than the previously proposed calcium ion (M. Chabre, Grenoble). It is proposed that rhodopsin floats in a fluid membrane and, on photoexcitation, interacts with an enzyme chain controlling cyclic GMP levels. There is evidence that the site of interaction of rhodopsin with membrane enzymes is remote from the retinal itself, which implies that there is an overall conformational change of the whole rhodopsin molecule. Support for this picture comes from the findings of H. Shichi and R.L. Somers (National Eye Institute) that purified rhodopsin illuminated *in vitro* activates a purified GTPase; this results from formation of a protein–rhodopsin complex and does not require a membrane environment.

Another important reversible photoreaction used by biological systems for sensory purposes is the **phytochrome system** of plants. H. Smith (University of Leicester) discussed the role of phytochrome in the natural environment. Light penetrating a plant canopy is enriched in far-red wavelengths (700–750 nm), because of absorption of other wavelengths by chlorophyll. This alters the phytochrome photoequilibrium, thus permitting the plant to detect the amount of canopy shade, and to react accordingly — either by growth out of the shaded area or by changing leaf and chloroplast development and metabolism to adapt to the shade regime. However, it is still not clear whether this detection depends on the ratio of the red and far-red forms of phytochrome or on the amount of the far-red form present.

Extremely sensitive **bioluminescence assays** were described by A.M. Michelson (Institut de Biologie Physico-Chimique, Paris), based upon the fact that clam luciferases behave like peroxidases, producing superoxide radical anion, which then causes the clam luciferin to luminesce. Such systems permit estimation of femtogramme quantities of peroxide, and exquisitely sensitive estimations of hydrogen peroxide, superoxide radical anion, and enzymes like peroxidase and superoxide dismutase. In enzyme assays, turnover of the enzyme provides an effective amplification of the light. Such techniques have also been used for estimation of antibodies bound either to such enzymes or to luciferases.

In the area of basic mechanisms in **ultraviolet photobiology**, progress has been made in the study of lethal effects of near-UV radiation (300–380 nm), which is present in sunlight at the surface of the Earth. M.J. Peak and J.G. Peak (Argonne National Laboratory) have shown that there is a 365 nm maximum in the action spectrum for induction of single-strand breaks in purified bacterial transforming DNA irradiated *in vitro*. These breaks are primarily true breaks (as opposed to alkali-labile bonds) and generally require oxygen; they might explain the small plateau observed at this wavelength in action spectra for killing of *E.coli* in air. However, work by A.G. Miguel and R.M. Tyrrell (Federal University of Rio de Janeiro) indicates that single-strand breaks are very readily repaired and are therefore probably not the major lethal lesion. Similar arguments have previously eliminated pyrimidine dimers from consideration. Thus, the primary near-UV lethal lesion in wild-type bacteria remains to be identified. Whatever the damage, one of its first manifestations appears (S.H. Moss and K.C. Smith, Stanford University School of Medicine) to be an altered cell membrane, which makes a significant contribution to the killing of cells plated on minimal medium.

An inducible repair system for near-UV lethal damage in *E. coli* has been found by J. Peters and J. Jagger (University of Texas at Dallas). The system accounts for a large fraction of lethality in actively growing cells, and is distinct from the far-UV-induced SOS system. In related work, S. Menezes and R.M. Tyrrell have shown that repair systems inducible by far-UV (200–300 nm) radiation play a relatively minor role in the repair of near-UV damage.

Photomedicine was much in evidence reflecting a maturation of photobiology in its application to medical problems. Of great interest were developments in so-called PUVA therapy, which involves the treatment of skin lesions, such as psoriasis, by administering the dye *P*soralen (orally) and then irradiating with *UV-A* light (near-UV light in the range 320–380 nm). This therapy has been extremely effective in producing clearance and remissions. Unfortunately, there is concern that long term PUVA may act as a promoter of non-melanoma skin cancer, although this hazard has not prevented some 80,000 patients in all parts of the world from utilizing the therapy because the disease itself is so disfiguring. P. Vigny, D. Averbeck and co-workers (Institut Curie, Paris) have now found that a psoralen derivative, 3-carbethoxypsoralen, does not produce interstrand DNA cross-links, in contrast to the commonly used 8-methoxypsoralen, and the new compound shows therapeutic activity in mice without exhibiting carcinogenesis. Additional new isopsoralen derivatives that produce only monofunctional addicts in DNA have been synthesized by the Padua University group of G. Rodighiero. This is a happy development which opens the way to possible broad use of light-and-chemical treatments (photochemotherapy) of skin disorders.

A second area of great medical interest concerns the immunological rejection of UV-induced **skin cancers**, studied by M.L. Kripke and co-workers (Frederick Cancer Research Center, Maryland). Many UV-induced squamous carcinomas and fibrosarcomas are immunologically rejected when transplanted to normal syngeneic recipients, but grow progressively in immunologically deficient animals or animals that have been UV irradiated (including the animals in which they were originally produced). Kripke and co-workers have found that antigens produced during UV irradiation of the skin trigger the production of suppressor T lymphocytes, which in turn prevent an immunological rejection of the tumour. Langerhans cells (macrophage-like cells in the skin) may be involved in the initial events in this process. K. Wolff (Innsbruck University) pointed out that Langerhans cells show the highest susceptibility to UV damage of all skin cells, and that UV

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0028-0836/81/080636-02$01.00

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abolishes their immunological surface markers and abrogates their capacity to stimulate T-lymphocyte proliferation. E.C. DeFabo and F.P. Noonan (Kripke's group) have found that the wavelength maximum for depression of contact sensitivity in mouse skin is at about 270 nm, which may reflect absorption by urocanic acid, a metabolite of histidine, which accumulates in the skin where it cannot be further metabolized. Urocanic acid, incidentally, has been proposed by J.A. Parrish (Massachusetts General Hospital) to act as a natural photoprotection (light-screening) agent, which is partially removed when skin is washed, explaining an observation first made by Jacopo Beccari in 1744 that his hands fluoresced better in sunlight after being washed.

Finally, a disturbing medical development is the recent rise in the incidence of malignant melanoma: world mortality rates have approximately doubled in the last fifteen years. It is not generally recognized that skin cancers are the most common malignant tumours of man. Although the overall mortality is less than 5%, malignant melanomas show about a 40% mortality, and they represented about 1% of all cancer deaths in 1970. The recent increase in melanoma incidence is generally attributed to increased sun-exposure habits, although it is possible that there is some kind of synergism between UV radiation and environmental toxic chemicals, as in the observations of P.D. Forbes and co-workers (Temple University Health Sciences Center) that concurrent exposure to retinoic acid and UV stimulates tumour development. Effective topical sunscreens are thus of great importance, and the need for international standardization of sunscreen efficacy was discussed in a Round-Table held at the congress.

Photobiology has become a very active discipline that is now rapidly maturing. Sophisticated physical and chemical techniques are being applied, and significant advances in fundamental knowledge are being made. It is playing an increasingly important role in medicine. All this comes as no surprise to the photobiologists, who share the view of Isaac Asimov that "The Earth is bathed in light from the Sun, and one could scarcely think of a more important single fact than that." □

Antisickling drugs and water structure

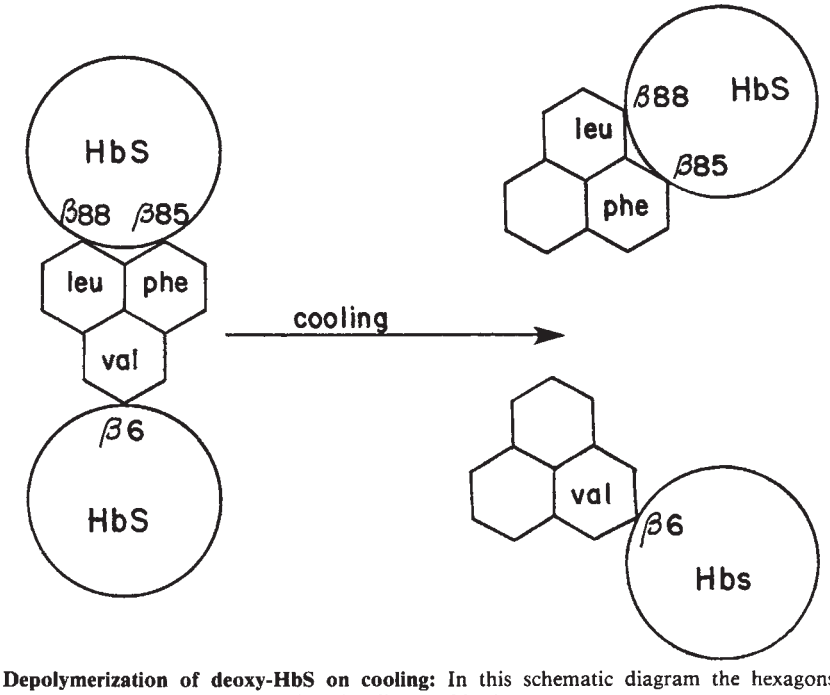
from Ruth and Reinhold Benesch

In sickle cell anaemia the substitution of a valine for a glutamic acid in the β chain of the normal haemoglobin molecule causes a polymerization of the deoxygenated sickle cell haemoglobin. In order to design drugs to prevent aggregation of the sickle cell haemoglobin (HbS) an understanding of the molecular mechanisms involved is obviously required[1,2]. Stillinger's[3] recent article, *Water Revisited,* has led us to suspect that the hydrophobic class of sickling inhibitors may act on water structure rather than by stereospecific interaction with HbS itself. Stillinger points out the analogy between hydrophobic interaction and the anomalous behaviour of supercooled water. Water is described as a random three-dimensional network of hydrogen bonds in which are embedded relatively bulky, unstrained hydrogen bond polyhedra. The concentration of these polyhedra increases as the temperature is lowered. The attractive force between them and their consequent 'clumping' is responsible for some of the unusual properties of supercooled water. When a non-polar solute is dissolved in water, the random network of hydrogen-bonded water molecules reorganizes around it to form very similar bulky convex polyhedra which only differ from the empty ones found in supercooled water by the presence of the hydrophobic molecule in their centre. The attraction between these 'filled' convex polyhedra provides the physical basis for the 'hydrophobic bond'.

A characteristic feature of hydrophobic interactions is their unusual temperature dependence, that is, they become weaker as the temperature is lowered. This can readily be explained in terms of the picture provided by Stillinger. Since lowering the temperature will increase the concentration of empty polyhedra, the probability of a filled one interacting with an empty one, rather than with another filled one, will increase and the attraction between the hydrophobic residues will weaken with decreasing temperature.

When sickle cell haemoglobin polymerizes it forms a gel that 'melts' as the temperature is lowered. Crucial side-to-side contact between strands of HbS molecules in the crystal (and therefore presumably also in the fibres formed by the haemoglobin) was shown to exist between valine β6 of one molecule and phenylalanine β85 and leucine β88 of the neighbouring one[4]. If this bond can now be regarded as the result of the attractive forces between hydrogen bond polyhedra containing these residues (see the figure), then it should be broken as the temperature is lowered because of competition by the empty water polyhedra as their concentration increases with decreasing temperature.

An analogous mechanism would also account for the well documented inhibition of HbS polymerization by a variety of compounds containing hydrophobic residues[5-9]. There is no clearcut relationship between the structure of these inhibitors and the known geometry of the sites containing valine β6 and phenylalanine β85



Depolymerization of deoxy-HbS on cooling: In this schematic diagram the hexagons represent the hydrogen bond polyhedra discussed in the text.

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0028-0836/81/080637-02$01.00

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and leucine β88. Thus, for example, the D isomeric forms of aromatic amino acids are as active as the L isomers[10]. This supports the hypothesis that the inhibition of polymerization by these compounds is brought about by competition between the water polyhedra filled with the small hydrophobic molecules with those attached to the protein, rather than a direct interaction between the hydrophobic residues and the protein which should be stereospecific. This picture would also explain why attempts to increase the effectiveness of hydrophobic antisickling agents by structural modification have met with relatively little success. □

1. Schechter, A. N. *Hemoglobin* **4**, 335 (1980).
2. *Development of Therapeutic Agents for Sickle Cell Disease INSERM Symp.* (eds Beuzard, Y., Hercules, J. & Rosa, J.) (Elsevier, Amsterdam, 1979).
3. Stillinger, F. H. *Science* **209**, 451 (1980).
4. Wishner, B. C., Ward, K. B., Lattman, E. E. & Love, W. E. *J. molec. Biol.* **98**, 179 (1975).
5. Elbaum, D., Nagel, R. L., Bookchin, R. M. & Herskovits, T. T. *Proc. natn. Acad. Sci. U.S.A.* **71**, 4718 (1974).
6. Ross, P. D. & Subramanian, S. *Biochem. biophys. Res. Commun.* **77**, 1217 (1977).
7. Votano, J. R., Gorecki, M. & Rich, A. *Science* **196**, 1216 (1977).
8. Kubota, S. & Yang, J. T. *Proc. natn. Acad. Sci. U.S.A.* **74**, 5431 (1977).
9. Behe, M. J. & Englander, S. W. *Biochemistry* **18**, 4196 (1979).
10. Noguchi, C. T. & Schechter, A. N. *Biochemistry* **17**, 5455 (1978).

Fifty years of Slough's ionosphere

from Henry Rishbeth

ON 11 January 1981 the 'noon run' of the Slough ionosonde marked with due ceremony the fiftieth anniversary of routine ionospheric sounding. An ionosonde is a kind of swept-frequency radar and radar developed from the experimental studies of the ionosphere that were conducted at Slough and elsewhere in the twenties and thirties. This work followed from the basic experiments of 1924–25 in Britain (E. V. Appleton and M. A. F. Barnett *Nature* **115**, 333; 1925) and in the US (G. Breit and M. A. Tuve *Nature* **116**, 357; 1925) that conclusively proved the existence of radio-reflecting layers in the upper atmosphere. It was Watson-Watt at the Radio Research Station (RRS), Slough (now the SRC Appleton Laboratory) who coined in 1926 the term 'ionosphere' (G. W. Gardiner *Nature* **224**, 1096; 1969). Years later, the output of analysis and interpretation based on Slough data gave rise to the unofficial definition of the ionosphere as 'the part of the atmosphere vertically above Slough'.

To promote scientific understanding of the ionosphere and to meet the growing needs of radio communications, systematic data were needed. RRS publications contain some h'–f data on apparent reflection heights (h') at different radio frequencies (f) obtained in 1930. However, the most important data on the ionospheric layers (E, F1 and F2 layers, respectively at heights of around 100, 180 and 250–350 km) are their 'penetration' or 'critical' frequencies (fE, fF1, fF2) as measured by a swept-frequency sounder. The series of critical frequency data began at noon on 11 January 1931 (a Sunday, like its fiftieth anniversary) and has continued with few breaks to the present day. Initially only fE was recorded, on ninety days in 1931. Tabulated fF2 data start on 19 December 1931, but it was not until 20 September 1932 that the long series of daily values was initiated, the first 'noon run' for which all three parameters fE, fF1, fF2 are preserved being on 23 September 1932. Hourly soundings — which are now made routinely — were made only for three 24-hour periods during 1931; from these data it appears that the initial value at 1200 GMT on 11 January 1931, namely 2.1 MHz, may be about 15–20 per cent low in comparison with neighbouring values. Furthermore, it was then also possible to record fE at night to values as low as 0.7 MHz: this could not be done nowadays with normal ionosondes in England, owing to the prevalence of radio broadcast interference (R. W. Smith, personal communication). Today, data from the Slough ionosonde are distributed to numerous users on schedules to meet their particular needs, which may include long-term 'predictions' of ionospheric conditions or short-term 'forecasts' of disturbances. Some users, including the BBC, Marconi's and Jodrell Bank, receive noon 'critical frequency' data by telex each working day. It is interesting that the largest value of fF2 ever recorded at Slough occurred in the present solar cycle, namely 17.2 MHz at 1300 and 1400 GMT on 15 December 1979.

Other long series of ionospheric data have been produced at Huancayo (Peru), Watheroo (Western Australia) and Washington (DC). By the start of the International Geophysical Year of 1957–58 about 140 ionospheric observatories were in operation, many of which have continued to this day, thereby creating global data sequences extending over two sunspot cycles. This well organized scientific resource is available to researchers through the World Data Centre at Slough, and corresponding Centres in Boulder (Colorado), Moscow and Tokyo, established at the start of the IGY.

What of the future? Following the merger with the SRC Rutherford Laboratory, Appleton Laboratory work is being transferred away from Slough. Current thinking is that an ionosonde should remain permanently in the Slough area. Possibilities being considered for the future are the automatic read-out of data to the merged laboratories' site at Chilton, with dial-up access by users both to past data and to the up-to-the-hour state of 'the part of the atmosphere vertically above Slough'.

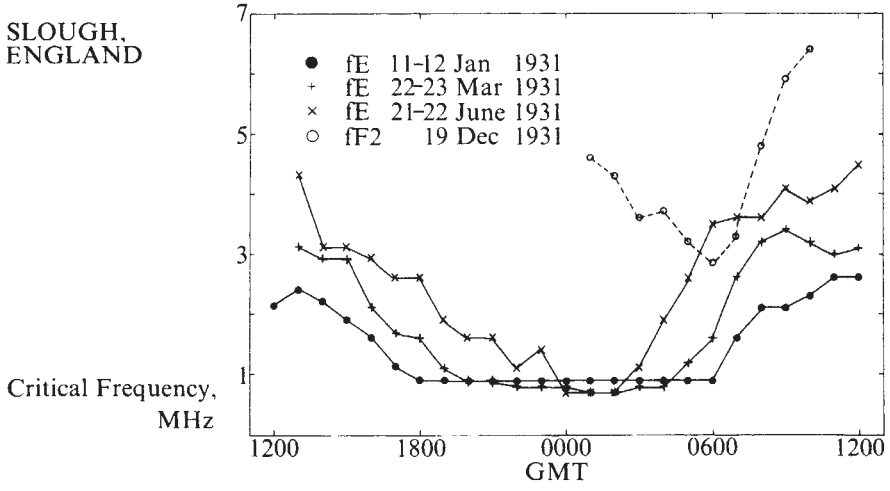

Fig. The four sequences of hourly critical frequency data recorded at Slough (51½°N, ½°W) during 1931
The values refer to the 'ordinary' magnetoionic component. Some of the fluctuations in fE are probably due to sporadic E.

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0028-0836/81/080638-01$01.00
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ARTICLES

Stable isotope composition of benthic foraminifera from the equatorial Pacific

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When ranked according to deviation from isotopic equilibrium, deep-sea benthic foraminifera exhibit no correlation between the rankings for oxygen and for carbon. Both rank sequences are stable from east to west in the equatorial Pacific (fertility-independent) and from Glacial to post-Glacial (time-independent). Records for the past 15,000 yr suggest marked changes in deep-sea circulation during this time interval.

STABLE isotopic compositions of benthic foraminifera reflect bottom water conditions. The $^{18}O/^{16}O$ ratio of the calcium carbonate in their shells is strongly dependent on the temperature and oxygen isotope composition of the ambient water, whereas the $^{13}C/^{12}C$ ratio is related to the carbon isotopic composition of the total dissolved inorganic carbon. Oxygen isotopic changes in benthic foraminifera from deep-sea sediments, first used in Emiliani[1,2] to reconstruct bottom water changes, have been used to reconstruct Pleistocene variations in Atlantic bottom water temperatures[3,4]. But changes in the carbon isotopic composition of benthic foraminiferal tests can also aid palaeoceanographic interpretations[5].

Although the initial assumption that all deep-sea benthic foraminifera deposit their shells in equilibrium with seawater[6] is incorrect[7–10], it is thought that for oxygen isotopes, deviation from equilibrium is small in some species[11,12], and that more substantial deviations of other species are constant and can be corrected for by adding a constant value to the measured $^{18}O/^{16}O$ ratio[5,13–19]. We show here that the ranges of variation within individual species and of isotopic differences between species are of the same order of magnitude as isotopic changes commonly recorded in Pleistocene sediments. Thus, our results impose limits on the significance of downcore isotope records based on single-species analyses, especially if small samples with few specimens are used.

Materials and methods

Benthic foraminifera from western and eastern equatorial Pacific sediments, recovered by box coring during Scripps Institution of Oceanography Expeditions Eurydice (ERDC, 1975) and Pleiades (PLDS, 1976) respectively, were analysed. Sedimentation patterns in the two areas have been described elsewhere[20,21]. They are characterized by relatively high sedimentation rates and high carbonate content (Table 1). Carbonate content in the east (PLDS) is lower than in the west (ERDC), largely because of high silica content in the eastern high fertility area. The higher fertility in the east is also reflected by the rate of sedimentation and a high ratio of *Neogloboquadrina dutertrei* to *Pulleniatina obliquiloculata* (Table 1), the former planktonic species being associated with upwelling[22]. For the present study, dissolution effects are not important above 3,000 m depth, and probably did not affect our results significantly, even in the two deepest cores.

We sampled the surface sediments in these box cores as well as a deeper level in Box-core ERDC-92 dated at 16,000–17,000 ^{14}C yr (ref. 23). All benthic foraminifera larger than 250 μm in each sample were selected and 150–400 specimens per sample were thus isolated. Of these specimens, 10 species were abundant enough to run replicate isotopic analyses (Table 2). Over 100 analyses for both oxygen and carbon were performed. Typically 4–7 specimens with a total weight of at least 0.5 mg were used for each analysis. Samples were cleaned ultrasonically to remove fine particles from inside the foraminiferal tests. Isotopic analysis followed standard procedures[24]. Benthic foraminifera taxonomy followed that of Culp[25].

In Core ERDC-92 foraminifera were analysed from discrete size fractions (149–250 μm, 250–420 μm and >420 μm). No correlation of isotopic composition with size was detected, however, in contrast to the well-established size effect on isotopic composition in planktonic foraminifera[26].

Species abundances

The two most common benthic species in samples shallower than 3,000 m are *Planulina wuellerstorfi* and *Oridorsalis umbonatus*, which each comprise 10–20% of the total number of individuals in the calcareous benthic assemblage. Below 3,000 m, these species and *Nuttalides umbonifera* dominate the benthic assemblages (ERDC-135 and PLDS-66). Here *N. umbonifera* represents 12–13% of the calcareous benthic fauna, whereas it is absent in the shallower samples.

All other species analysed are less common and generally constitute <5% of the calcareous benthic fauna. *Uvigerina*, which is rare (<2%) or absent in all ERDC samples, is common in the PLDS-66 sample, making up 15% of the calcareous benthic fauna. The higher productivity of surface waters in the eastern region is presumed to cause an increased supply of food to the underlying benthos. Therefore we suggest that the abundance of *Uvigerina* indicates a region of high fertility. This

Table 1 Core location and sample material

Box core no.	Lat	Long	Water depth (m)	Depth in core (cm)	Age	Sedimentation rate* (cm kyr^{-1})	$CaCO_3$	dut./Pul.†
ERDC-92	2°13.5′S	156°59.9′E	1,598	0–1.5	Holocene	1.6	90%	<1
ERDC-92	2°13.5′S	156°59.9′E	1,598	20–30	Glacial	1.8	86%	5
ERDC-123	0°01.3′S	160°24.9′E	2,948	0–1	Holocene	1.8	84%	<1
ERDC-135	0°52.5′N	160°59.6′E	3,509	0–2	Holocene	1.4	85%	<1
PLDS-66	0°56.6′N	104°6.1′W	3,496	0–2	Holocene	2.0	72%	72

* Derived from ^{14}C dates (ERDC-92, ref. 23; ERDC-123, 135, ref. 32; PLDS-66, unpublished data).
† Ratio of *N. dutertrei* to *P. obliquiloculata* (>149 μm).

0028–0836/81/080639—05$01.00
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Table 2 Isotopic composition of various benthic foraminiferal species

Groups	Species	(1) ERDC-92, Holocene		(2) ERDC-92, Glacial	(3) ERDC-123, Holocene		(4) ERDC-135, Holocene		(5) PLDS-66, Holocene	
a $\delta^{13}O$										
"Low ^{18}O"	*Nuttalides umbonifera*	—		—	—		3.00 (3) *r* 0.84	−0.60	2.43 (3) *r* 0.30	−1.17
	Laticarinina pauperata	2.65 (6) *r* 0.84 ± 0.29	−0.55	3.10 (2) *r* 0.67	2.89 (1)	−0.61	3.20 (1)	−0.40	2.69 (3) *r* 0.46	−0.91
	Planulina wuellerstorfi	2.69 (12) *r* 1.12 ± 0.42	−0.51	3.13 (5) *r* 1.14 ± 0.51	2.66 (3) *r* 0.21	−0.84	3.21 (2) *r* 0.45	−0.39	2.50 (4) *r* 0.06 ± 0.04	−1.10
	Favocassidulina favus	—		—	2.98 (1)	−0.52	3.09 (3) *r* 0.46	−0.51	—	
"High ^{18}O"	*Oridorsalis umbonatus*	3.23 (8) *r* 1.58 ± 0.49	+0.03	3.99 (3) *r* 0.49	3.34 (3) *r* 0.22	−0.16	3.66 (3) *r* 0.65	+0.06	3.33 (3) *r* 0.46	−0.27
	Siphouvigerina interrupta	3.25 (2) *r* 0.36	+0.05	3.99 (2) *r* 0.44	—		—		—	
	Cassidulina subglobosa	3.33 (4) *r* 1.01 ± 0.43	+0.13	4.55 (4) *r* 0.27 ± 0.12	3.33 (1)	−0.17	3.70 (1)	+0.10	3.18 (3) *r* 0.31	−0.41
	Quinqueloculina auberiana	3.21 (2) *r* 0.07	+0.01	4.30 (2) *r* 0.07	—		—		—	
	Uvigerina spp.	—		—	—		—		3.16 (4) *r* 0.64 ± 0.27	−0.44
	Hoeglundina elegans	4.04 (5) *r* 0.67 ± 0.27		4.79 (4) *r* 0.70 ± 0.29						
Estimated equilibrium*		3.2		—	3.5		3.6		3.6	
b $\delta^{13}C$										
"Super low ^{13}C"	*Oridorsalis umbonatus*	−0.53 (8) *r* 0.53 ± 0.18	−1.93	−0.75 (3) *r* 0.79	−0.89 (3) *r* 0.36	−2.29	−0.93 (3) *r* 0.29	−2.43	−1.17 (3) *r* 0.35	−2.37
	Favocassidulina favus	—		—	−0.66 (1)	−2.06	−0.61 (3) *r* 0.23	−2.11	—	
"*Low* ^{13}C"	*Uvigerina* spp.	—		—	—		—		−0.63 (4) *r* 0.48 ± 0.21	−1.83
	Siphouvigerina interrupta	−0.14 (2) *r* 0.22	−1.54	−0.47 (2) *r* 0.07	—		—		—	
	Cassidulina subglobosa	0.04 (3) *r* 0.37	−1.36	−0.49 (4) *r* 0.47 ± 0.22	−0.59 (1)	−1.99	−0.56 (1)	−2.06	−0.61 (3) *r* 0.36	−1.81
"Medium ^{13}C"	*Laticarinina pauperata*	−0.1 (6) *r* 1.41 ± 0.48	−1.41	−0.20 (2) *r* 0.25	−0.18 (1)	−1.58	−0.28 (1)	−1.78	−0.21 (3) *r* 0.09	−1.41
	Nuttalides umbonifera	—		—	—		−0.23 (3) *r* 0.70	−1.73	−0.18 (3) *r* 0.33	−1.38
"High ^{13}C"	*Planulina wuellerstorfi*	0.40 (12) *r* 0.49 ± 0.12	−1.00	0.45 (5) *r* 0.33 ± 0.15	0.24 (3) *r* 0.28	−1.16	0.07 (2) *r* 0.02	−1.43	0.19 (4) *r* 0.39 ± 0.18	−1.01
	Quinqueloculina auberiana	0.64 (2) *r* 0.25	−0.76	1.13 (2) *r* 0.16	—		—		—	
	Hoeglundina elegans	2.13 (5) *r* 0.29 ± 0.11		2.14 (4) *r* 0.32 ± 0.16	—		—		—	
Estimated equilibrium†		1.4		—	1.4		1.5		1.2	

All specimens are >250 μm except *S. interrupta*, which is from the 149–250 μm size fraction. In each column the first number to the left is the mean δ value with respect to ‰ PDB; the number in parentheses is the number of analyses followed by the range *r* and by the standard deviation if more than three analyses were made; the last number, ro the right, is the $\Delta\delta$ value (the mean δ value minus the estimated equilibrium value).

* For present-day conditions the $^{18}O/^{16}O$ composition of calcite at equilibrium with ambient-bottom water was calculated using a palaeotemperature equation[44] and estimated temperatures[45] of 3°, 1.8°, 1.5° and 1.5 °C for ERDC 92, 123, 135 and PLDS 66, respectively. The isotopic composition of the bottom water was assumed to be −0.4 (PDB) (ref. 46).

† Present-day equilibrium $^{13}C/^{12}C$ composition of equilibrium calcite was estimated for the four core sites using reported fractionation relationships between calcium carbonate and dissolved bicarbonate[47]. Ambient $\delta^{13}C$ (PDB) values for bicarbonate were calculated as 0.15, 0.18, 0.25, and 0.25 for cores ERDC 92, 123, 135 and PLDS 66, respectively, from apparent oxygen utilization (AOU) to $\delta^{13}C$ relationships[13]. AOU values were obtained from reported temperature, salinity and dissolved O_2 data[45,48].

assessment is supported by its decreasing abundance in Box-Core ERDC-123 from Glacial to Holocene sediments (from 6% of the benthic calcareous assemblage to <1%) in parallel to other trends commonly associated with decreasing fertility: (1) a decreasing sedimentation rate (from 3 to 1.8 cm kyr^{-1}); (2) a decreasing *N. dutertrei/P. obliquiloculata* ratio (from >1 to 0.2); and (3) a decreasing benthic foram number (from ~30 to 10 benthic foraminifera larger than 250 μm g^{-1} of dry sediment).

Oxygen isotopes

Results of the oxygen analyses are given in Table 2*a*. Species are listed in the order of deviation from estimated equilibrium values, which for $\delta^{18}O$ in present day conditions range from 3.2 to 3.6‰, depending on the water depth of the core location (see Table 2*a*). Our comparisons are between species, between periods (ERDC-92, Holocene and Glacial), and between areas of low fertility in the west (ERDC) and high fertility in the east (PLDS). The disequilibrium ranking (greatest disequilibrium first) consists of two groups: 'low ^{18}O' and 'high ^{18}O'. The low ^{18}O group includes *N. umbonifera, L. pauperata, P. wuellerstorifi*, and *Favocassidulina favus*, which are clearly depleted in ^{18}O relative to equilibrium by around 0.5‰ in the ERDC samples. The high ^{18}O group contains *O. umbonatus, Siphouvigerina interrupta, Cassidulina subglobosa, Quinqueloculina auberiana* and *Uvigerina* spp., which have oxygen values close to equilibrium (for *Uvigerina* see ref. 5). *Hoeglundina elegans* is a separate case: its shell is aragonitic[27–29]. The equilibrium values for aragonite are not known for these low temperatures.

The grouping into low ^{18}O and high ^{18}O seems unrelated to time (column 1 compared with column 2) and with fertility (columns 1, 3, 4 compared with column 5). From the $\Delta\delta^{18}O$ values (the mean δ values minus the estimated equilibrium values) in Table 2*a* and Fig. 1*a*, it seems that there is a fertility effect on isotopic composition, because the degree of disequilibrium for oxygen increases from west to east. Compared with the eastern core (PLDS 66), the five benthic species shown for the western cores (ERDC) all contain a higher $^{18}O/^{16}O$ ratio and are closer to estimated equilibrium values. Moreover, between the western cores an increase in disequilibrium with increasing sedimentation rate is indicated, which in turn is related to fertility.

The large range in most of the replicate analyses makes it impossible to confirm that the amount of change in disequilibrium with fertility is species dependent.

Carbon isotopes

Results of the carbon analyses are given in Table 2*b*. Again, species are listed in the order of deviation from estimated equilibrium values, which range in $\delta^{13}C$ from 1.2 to 1.5‰, depending on the apparent oxygen utilization (AOU) in the water. The AOU indicates how much oxygen has been used to oxidize organic matter to produce additional CO_2 (with a $\delta^{13}C$ near −20‰)[26,30]. The ranking (greatest disequilibrium first) consists of four groups: 'super low ^{13}C', 'low ^{13}C', 'medium ^{13}C', and 'high ^{13}C'. The super low ^{13}C group includes *O. umbonatus* and *F.favus*, the low ^{13}C group includes *Uvigerina* spp., *S. interrupta*, and *C. subglobosa*, the medium ^{13}C group includes *L. pauperata* and *N. umbonifera*, and the high ^{13}C group is made up of *P. wuellerstorfi* and *Q. auberiana*. Again, the aragonitic *H. elegans* is anomalous, with very high $\delta^{13}C$ values.

The carbon ranking's apparent independence of the oxygen ranking, may indicate that at least two mechanisms of fractionation are involved which operate more or less independently to produce the 'vital effects' in oxygen and carbon isotopic compositions. As was shown for oxygen, ranks apparently are time- and fertility-invariant (columns 1 compared with 2; and 1, 3, 4 compared with 5). Comparison of the degree of disequilibrium for carbon in the western area with that in the eastern area (Fig. 1*b*) shows that the change in disequilibrium is not as coherent as it seems for oxygen. However, most of the $\delta^{13}C$ values are closer to equilibrium in the eastern core than in the western cores—a reverse trend to the oxygen trend.

Interspecific and intraspecific ranges

In the Holocene samples, interspecific $\Delta\delta$ ranges are near 1‰ or less for oxygen and more than 1‰ for carbon (Table 2, Fig. 1). Intraspecific (within species) ranges in both $\delta^{18}O$ and $\delta^{13}C$ are commonly of the order of several tenths of 1‰ (also see ref. 10). Maximum intraspecific ranges within single samples are near 1.5‰ for both $\delta^{18}O$ and $\delta^{13}C$. Isolated determinations should, therefore, be interpreted with caution as indicators of changes in either ambient conditions or vital effects.

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From Fig. 1, the relative position of *O. umbonatus* data with respect to the general trend of the other three species suggests that *O. umbonatus* may be less affected by fertility changes, although more data are necessary to confirm this. Nevertheless, our data do not support the conclusion of Belanger *et al.*[31] that *P. wuellerstorfi* is a more reliable indicator of bottom water than *O. umbonatus*.

Oxygen isotope record of the past 15,000 yr

The relationships between the isotopic signals of various deep-sea benthic foraminifera are further illustrated by a 15,000-yr record, based on closely spaced samples of Box Core ERDC-123 (Fig. 2). The carbon-14 stratigraphy of this core[32] (see Fig. 2, lower scale) shows that sedimentation was continuous. The sedimentation rate is ~3 cm kyr^{-1} from the base of the core to ~20 cm, and changes rather abruptly to 1.8 cm kyr^{-1} above 20 cm in the core, at ~10,000 yr BP. Sharply lower sedimentation rates for the post-Glacial, compared with the Glacial, have been reported for this area[23,33].

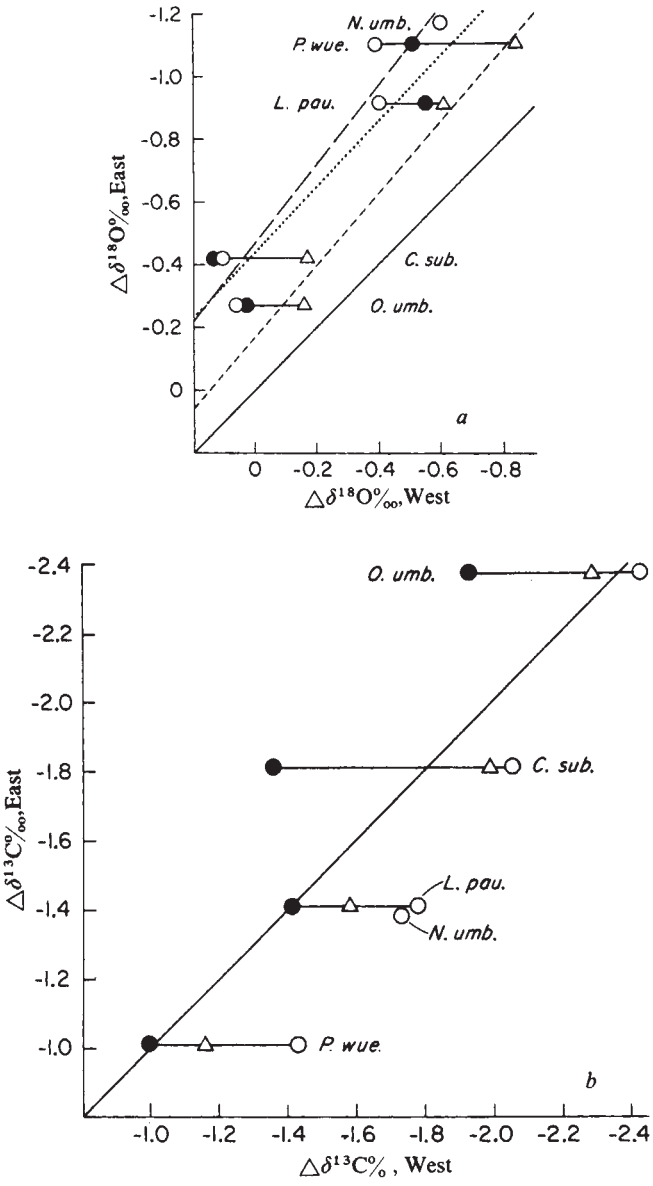

Fig. 1 Comparison of the degree of disequilibrium in recent benthic foraminifera for oxygen (*a*) and carbon (*b*) between west and east in the equatorial Pacific. $\Delta\delta$ values are from Table 2. Western cores: ERDC-92 (●), ERDC-123 (△), ERDC-135 (○); eastern core: PLDS-66. *N. umb.*, *Nuttalides umbonifera*; *P. wue.*, *Planulina wuellerstorfi*; *L. pau.*, *Laticarinina pauperata*; *C. sub.*, *Cassidulina subglobosa*; *O. umb.*, *Oridorsalis umbonatus*. The solid 45° line corresponds to an equal disequilibrium between west and east. The other lines are the best calculated fits for the oxygen isotopic data (long dashed line: ERDC-135, correlation coefficient of the fit, $r = 0.96$; dotted line: ERDC-92, $r = 0.93$; short dashed line: ERDC-123, $r = 0.99$).

The oxygen isotope record of shallow-living (*Globigerinoides sacculifer*) and deep-dwelling (*P. obliquiloculata* and *N. dutertrei*) planktonic foraminifera shows the familiar change from Glacial to post-Glacial values for this area[25]. The maximum rate of change occurs between 21 and 29 cm (13,000–10,000 ^{14}C yr). Three factors are involved in producing the change: (1) introduction of meltwater into the world ocean; (2) possible incomplete mixing of this meltwater; and (3) global warming. The relative importance of these factors to the rate of change is not clear, but we assume that the first can account for a total change of near 1‰, the second for a transient change of near 0.5‰, the third for about 0.4‰[24,34,35]. An additional factor, mixing by benthic organisms, decreases the recorded rate of change[36–39].

The oxygen isotope stratigraphy of benthic foraminifera also shows the deglaciation change, but continuing beyond 21 cm until values reach a light δ^{18}O plateau around 15 cm (7,500 ^{14}C yr BP). The difference in the isotope records does not necessarily indicate a 'lead-lag' relationship such as has been postulated for various Pleistocene palaeoceanographic signals[40,41]. The fact that the abundance of the carrier particles for the isotope signals changes across the Glacial–Holocene boundary will by itself change the shape of the signal, as pointed out earlier[34]. As the relative abundance of the benthic carrier decreases markedly across the boundary, its isotopic signal may lag behind that of the planktonic through the effects of mixing because proportionally more benthic signal carriers (with high δ^{18}O values) are mixed upwards when compared with the planktonic signal carriers. Recent numerical experiments by W. H. Hutson (personal communication) have demonstrated the importance of mixing on such lead-lag phenomena.

The various benthic signals are reasonably parallel, only the one of *F. favus* crosses any of the others. The rapid increase in δ^{18}O values in all four species near 10 cm (5,000 ^{14}C yr BP) and the distinct trend to lighter values near the surface of the sediment are remarkable. (The uppermost sediment in this core is in fact ^{14}C young and not yet absorbed into the mixed layer[32].)

The range of the deglacial δ^{18}O change, including the early Holocene plateau, is greatest in *P. wuellerstorfi*, the most oxygen-disequilibrated species of the four. Its range is 2‰. The other species show ranges between 0.9 and 1.4‰, for an average of 1.2‰. The cause of the benthic oxygen isotope plateau between 5,500 and 8,500 ^{14}C yr BP is not immediately clear, but it is possibly associated with the period of the "Climatic Optimum"[42].

We suggest the following interpretation of the oxygen isotope record: (1) a lag in the effect of glacial melting on the bottom water composition, with bottom waters reaching Holocene values ~2,000 yr after the upper waters; (2) a period in which benthic foraminifera have abnormally low ^{18}O values because of a decrease in bottom water production, and hence warming of bottom waters, during the climatic optimum; and (3) an increase of bottom water production after the climatic optimum, with a concomitant rapid drop in abyssal temperatures with the onset of "Neoglaciation"[42]. Future work will show whether these suggestions are acceptable or not. Clearly, however, ice volume cannot be read directly from the benthic signals (as has been variously proposed), because of: (1) differences in amplitudes between species; (2) existence of transient events throughout deglaciation and the Holocene; (3) mixing effects; and (4) intraspecific variation in δ^{18}O.

Carbon isotope record of the past 15,000 yr

The carbon isotope stratigraphy of the deep-dwelling planktonic foraminifera *P. obliquiloculata* is remarkable for its lack of structure. Apparently we need not postulate significant variation in δ^{13}C of upper ocean waters during the period in question.

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The surface-dwelling *Globigerinoides sacculifer* shows a stepped signal: low values (1.65‰ ± 0.15) from 15,000 to 9,000 ^{14}C yr ago, high values (2.0‰ ± 0.2) since 9,000 yr BP. The change correlates with the previously mentioned changes in sedimentation rate and in benthic foraminiferal number. A distinct decrease in equatorial upwelling within the early Holocene, with concomitant stabilization of a ^{13}C-rich surface layer is suggested. A marked decrease in fertility from Glacial to post-Glacial time is also indicated by the δ^{13}C values of the fine sand fraction ('Fines'). Low δ^{13}C in this fraction is believed to indicate high metabolic activity[26], and such activity seems to decrease with time.

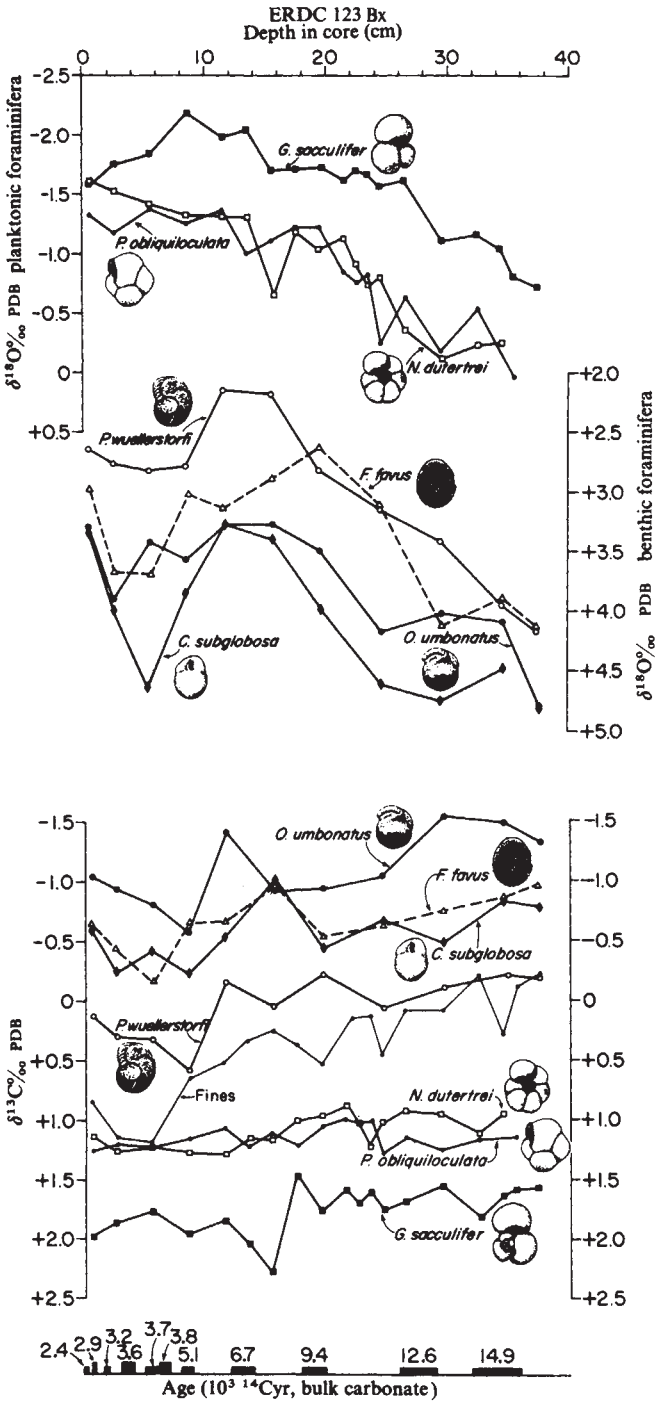

Fig. 2 Oxygen and carbon isotope stratigraphies in Box Core ERDC-123 for the shallow-living planktonic foraminifera *Globigerinoides sacculifer* (250–420 μm), the deep-dwelling ones *Pulleniatina obliquiloculata* and *Neogloboquadrina dutertrei* (250–420 μm) and the benthic species *Cassidulina subglobosa, Favocassidulina favus, Oridorsalis umbonatus* and *Planulina wuellestorfi* (>250 μm). Carbon isotope stratigraphy for the fine sand fraction ('Fines': 63–125 μm). Foram shells drawn by F. L. Parker (planktonics) and W. T. Coulbourn (benthics).

The decrease in fertility from Glacial to post-Glacial time is also reflected in an increase in δ^{13}C values of benthic foraminifera in post-Glacial sediments. Possibly the oxygen isotope plateau in the early Holocene is accompanied by a lightening of the ambient δ^{13}C values: if the plateau was partly due to warming, and hence incomplete mixing, isotopically light CO_2 should have built up. A CO_2 buildup would obscure a general δ^{13}C increase from lowered fertility, and shift the high benthic δ^{13}C values into the late Holocene. There is some indication in all four benthic signals that such a shift is indeed present, compared with the planktonic signal. Again, the effects of mixing on the signal, through changing signal carrier abundance may bias results in this direction.

Five of the benthic taxa analysed in this study (*C. subglobosa, F. favus, O. umbonatus, P. wuellerstorfi* and *Uvigerina* spp.) maintain the same relative positions in terms of δ^{18}O and δ^{13}C in Upper Miocene sediments[43], demonstrating that the relative intensity of vital effects in the various species tend to be retained over millions of years.

Conclusion

We have established the degree of disequilibrium precipitation in various deep-sea benthic foraminifera, and shown how this helps the interpretation of isotope record. Stratigraphies using multiple species are necessary for a more than superficial understanding of palaeoceanographic history, especially on a time scale of <10,000 yr. On this time scale, unfortunately, mixing processes disrupt the deep-sea record sufficiently to make stratiagraphic interpretations rather tentative, until more cores are studied and mixing processes are more quantitatively understood.

We thank Susan K. Culp, Richard F. Johnson, and Frances L. Parker for helpful discussions, and Thomas J. Walsh for assistance in collecting and sampling the box cores. We also thank Samuel M. Savin for constructive criticism. This study was supported by the NSF (Oceanography Section) grant OCE78-25587.

Received 2 July; accepted 2 December 1980.

1. Emiliani, C. *J. Geol.* **63,** 538–578 (1955).
2. Emiliani, C. *Science* **119,** 853–855 (1954).
3. Duplessy, J. C., Chenouard, L. & Villa, F. *Science* **188,** 1208–1209 (1975).
4. Streeter, S. S. & Shackleton, N. J. *Science* **203,** 168–171 (1979).
5. Shackleton, N. J. in *The Fate of Fossil Fuel CO_2 in the Oceans* (eds Andersen, N. R. & Malahoff, A.) 401–427 (Plenum, New York, 1977).
6. Shackleton, N. J. *Nature* **215,** 15–17 (1967).
7. Duplessy, J. C., Lalou, C. & Vinot, A. C. *Science* **168,** 250–251 (1970).
8. Vinot-Bertouille, A. C. & Duplessy, J. C. *Earth planet. Sci. Lett.* **18,** 247–257 (1973).
9. Vincent, E., Killingley, J. S. & Berger, W. H. *Geol. Soc. Am., Abstr. Prog.* **11,** 532–533 (1979).
10. Woodruff, F., Savin, S. M. & Douglas, R. G. *Mar. Micropaleontol.* **5,** 3–11 (1980).
11. Smith, P. B. & Emiliani, C. *Science* **160,** 1335–1336 (1968).
12. Shackleton, N. J. *Colloques Int. Cent. natn. Rech. scient.* **219,** 203–209 (1974).
13. Boersma, A. & Shackleton, N. J. *Init. Rep. DSDP* Leg **39,** 911–924 (1974).
14. Duplessy, J. C. in *Climatic Changes* (ed. Gribbin, J.) 46–67 (Cambridge University Press, 1978).
15. Pastouret, L., Chamley, H., Delibrias, G. & Duplessy, J. C. *Oceanol. Acta* **1,** 217–232 (1978).
16. Keigwin, L. D. Jr *Earth planet. Sci. Lett.* **45,** 361–382 (1979).
17. Keigwin, L. D. Jr, Bender, M. L. & Kennett, J. P. *Science* **205,** 1386–1388 (1979).
18. Shackleton, N. J. & Cita, M. B. *Init. Rep. DSDP* Leg **47,** 433–445 (1979).
19. Shackleton, N. J. & Opdyke, N. D. *Quat. Res.* **3,** 39–55 (1973).
20. Berger, W. H., Johnson, T. C. & Hamilton, E. L. in *The Fate of Fossil Fuel CO_2 in the Oceans* (eds Andersen, N. R. & Malahoff, A.) 543–567 (Plenum, New York, 1977).
21. Johnson, T. C., Hamilton, E. L. & Berger, W. H. *Mar. Geol.* **24,** 259–277 (1977).
22. Parker, F. L. & Berger, W. H. *Deep Sea Res.* **18,** 73–107 (1971).
23. Peng, T. H., Broecker, W. S. & Berger, W. H. *Quat. Res.* **11,** 141–149 (1979).
24. Berger, W. H. & Killingley, J. S. *Science* **197,** 563–566 (1977).
25. Culp, S. K. M.S. thesis, Univ. Hawaii (1977).
26. Berger, W. H., Killingley, J. S. & Vincent, E. *Oceanol. Acta* **1,** 203–216 (1978).
27. Bandy, O. L. *J. Sediment. Petrol.* **24,** 60–61 (1954).
28. Blackmon, P. D. & Todd, R. *J. Paleontol.* **33,** 1–15 (1959).
29. Vénec-Peyré, M. T. & Jaeschke-Boyer, H. *C.r. hebd. Séanc. Acad. Sci, Paris* **287,** 607–609 (1978).
30. Williams, D. F., Sommer, M. A. & Bender, M. L. *Earth planet. Sci. Lett.* **36,** 391–403 (1977).
31. Belanger, P. E., Curry, W. B. & Matthews, R. K. in *Oxygen and Carbon Isotopes in Foraminifera* (eds Berger, W. H., Bé, A. W. H. & Vincent, E.) (Elsevier, Amsterdam, in the press).
32. Krishnamurthy, R. V., Lal, D. Somayajulu, B. L. K. & Berger, W. H. *Proc. Ind. Acad. Sci.* 88A Pt. II, 3, 273–283 (1979).

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33. Berger, W. H. & Johnson, R. F. *Earth planet. Sci. Lett.* **41**, 223–227 (1978).
34. Berger, W. H., Johnson, R. F. & Killingley, J. S. *Nature* **269**, 661–663 (1977).
35. Vincent, E. & Berger, W. H. in *The Sea*, 7 (ed., Emiliani, C.) (Wiley, New York, in the press).
36. Berger, W. H. & Heath, G. R. *J. mar. Res.* **26**, 134–143 (1968).
37. Nozaki, Y., Cochran, J. K., Turekian, K. K. & Keller, G. *Earth planet. Sci. Lett.* **34**, 167–184 (1977).
38. Peng, T. H., Broecker, W. S., Kipphut, G. & Shackleton, N. in *The Fate of Fossil Fuel CO_2 in the Oceans* (eds Andersen, N. R. & Malahoff, A.) 355–373 (Plenum, New York, 1977).
39. Berger, W. H., Ekdale, A. A. & Bryant, P. F. *Mar. Geol.* **32**, 205–230 (1979).
40. Moore, T. C. Jr, Pisias, N. G. & Heath, G. R. in *The Fate of Fossil Fuel CO_2 in the Oceans* (eds Andersen, N. R. & Malahoff, A.) 145–165 (Plenum, New York, 1977).
41. Thompson, P. R. *J. Foram. Res.* **6**, 208–227 (1976).
42. Flint, R. F. *Glacial and Quaternary Geology* (Wiley, New York, 1971).
43. Vincent, E., Killingley, J. S. & Berger, W. H. *Mar. Micropaleonol.* **5**, 185–203 (1980).
44. Epstein, S., Buchsbaum, K., Lowenstam, H. & Urey, H. C. *Bull. geol. Soc. Am.* **64**, 1315–1326 (1952).
45. Reid, J. L. *Intermediate Waters of the Pacific Ocean*, 7–85 (The Johns Hopkins University Press, 1965).
46. Craig, H. & Gordon, L. I. in *2nd Conf., Stable Isotopes in Oceanographic Studies and Paleotemperatures* (ed. Tongiorgi, E.) 9–130 (Consiglio Nazionale delle Richerche, Rome, 1965).
47. Emrich, K., Ehhalt, D. H. & Vogel, J. C. *Earth planet. Sci. Lett.* **8**, 363–371 (1970).
48. Kroopnick, P. M., thesis, Univ. California, San Diego (1971).

Mouse liver and salivary gland α-amylase mRNAs differ only in 5′ non-translated sequences

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The sequence of the 1,773-nucleotide major and 1,806-nucleotide minor mouse liver α-amylase mRNAs differ only with respect to approximately 30 additional residues at the extreme 5′ terminus of the minor species. Comparison of the liver α-amylase mRNAs with their salivary gland counterpart reveals that these mRNAs share identical coding and 3′-noncoding sequences, but contain distinct 5′-terminal residues. These data suggest that all three mRNAs might be transcribed from the same gene.

To gain insight into the mechanisms which govern tissue-specific gene expression, we have investigated α-amylase mRNA biosynthesis in various mouse tissues. Early reports from other investigators indicated that α-amylase is produced in the pancreas, the salivary gland and the liver[1–3] and that the pancreatic protein differs from the other two with respect to antigenic and charge properties[4–9]. In our recent report of the primary structure of the α-amylase mRNAs which accumulate in the pancreas and the salivary gland, we found that the two mRNAs differ in nucleotide sequence, indicating that they are transcribed from different genes[10,11].

The liver α-amylase mRNA is less well characterized. Schibler *et al.*[10] showed that this mRNA is 100-fold less abundant and ~100 nucleotides longer than its salivary gland counterpart. However, the two species were found to have very similar sequences as indicated by their melting behaviour and resistance to S_1 nuclease digestion when part of α-amylase cDNA–mRNA hybrids. Restriction endonuclease digestion of cloned cDNAs made against liver and salivary gland α-amylase mRNA revealed no differences in cleavage sites for the various enzymes tested[12].

To elucidate structural relationships between α-amylase mRNAs produced in the salivary gland and liver in more detail, we have determined the nucleotide sequence of liver α-amylase mRNA. Here we report the intriguing finding that the liver and salivary gland α-amylase mRNAs encode identical proteins but differ in the terminal portion of their 5′-non-translated regions.

5′-terminal sequences account for the length difference between salivary gland and liver α-amylase mRNAs

Hybrids between salivary gland α-amylase cDNA and liver mRNA are resistant to S_1 nuclease along the entire length of the 1,600-nucleotide cDNA[10], suggesting that sequences which are responsible for the length difference between salivary gland and liver mRNAs reside at one or both ends of the liver species. To locate these extra sequences, we exploited the ability of RNase H to digest the RNA moiety of RNA–DNA hybrids[13–15]. The experimental approach is outlined in Fig. 1*a*. The 520-base pair *Eco*RI fragment of the cDNA clone pMSa104 (Fig. 1*b*), which contains translated sequences from the salivary gland α-amylase mRNA, was hybridized to polyadenylated RNA from pancreas, salivary gland or liver. Hybrids were then digested with RNase H and the products separated on a methyl mercury hydroxide agarose gel and transferred to diazotized (DMB) paper. The 5′ or 3′ RNase H-resistant RNA fragments were visualized by hybridization with appropriate 5′- or 3′-specific probes generated from the pMSa104 cDNA clone (Fig. 1*b*).

The autoradiographs shown in Fig. 1 indicate that sequences which account for the differences in size between the liver and salivary gland α-amylase mRNAs reside at the 5′ terminus. The results for the pancreas and salivary gland mRNAs (Fig. 1*c*) concur with those expected: only one major α-amylase mRNA accumulates in each of these two tissues and the salivary gland mRNA contains 80 more 5′-terminal noncoding residues than its pancreatic counterpart[10,11]. Two 5′-terminal liver RNA fragments are revealed in this experiment. Using the predicted sizes of the pancreas and salivary gland 5′-terminal fragments, we estimate that the two liver fragments are 490 and 520 nucleotides long. Densitometer tracings indicate that the larger fragment is present at 20% the level of the smaller one. For convenience, we will refer to these two liver RNAs as 5′-major and 5′-minor RNAs, reflecting their relative concentrations.

No apparent length differences are observed in 3′-terminal α-amylase mRNA fragments from the three tissues (Fig. 1*d*). However, an overexposure of the autoradiograph revealed a minor RNA fragment in both salivary gland and liver; Fig. 1*e* shows a longer exposure of the lane containing liver RNA. The size and abundance of this fragment indicate that it originates from a previously described minor α-amylase mRNA species (3′-minor RNA). Present at about 5% the level of total α-amylase mRNA in both tissues, the 3′-minor species contains major species sequences plus 237 additional nucleotides at its 3′ end[12].

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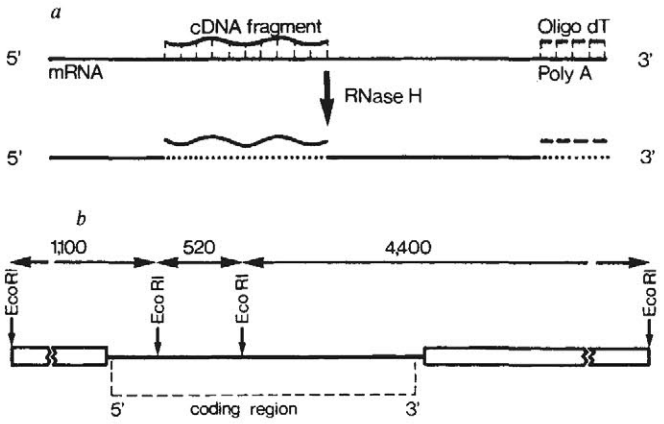
Sequence analysis of liver α-amylase mRNAs

To determine the liver α-amylase mRNA sequence, our strategy included elucidating the cDNA sequence. Double-stranded cDNA was synthesized against liver poly(A)$^+$ RNA and cloned in *E. coli*[12]. Due to the low abundance of α-amylase mRNA in this tissue (0.02% of poly(A)$^+$ RNA), only one recombinant plasmid (pMLa52) was obtained. Restriction endonuclease mapping of pMLa52 DNA suggested that it contained sequences complementary to the 3′-minor α-amylase mRNA; subsequent sequence analysis of residues representing the extreme 3′ terminus confirmed this interpretation[12]. Because the 3′-minor species is simply an extended version of the 3′-major liver α-amylase mRNA (see above), we could use pMLa52 DNA to determine sequences for both mRNAs.



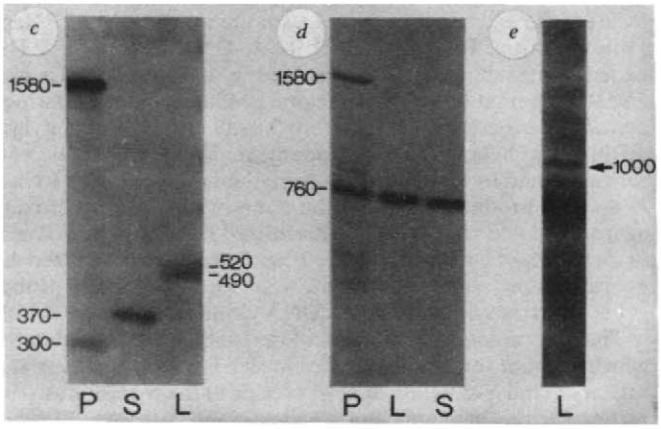


Fig. 1 Locating sequences which account for the length differences between liver, salivary gland and pancreas α-amylase mRNAs. *a*, the strategy used to generate 5′- and 3′-terminal fragments of α-amylase mRNA using RNase H. *b*, Diagram of pMSa104 DNA[10] linearized with respect to the pBR322 *Eco*RI site. Salivary gland α-amylase cDNA insert is represented as a line, pBR322 vector is boxed. Arrows delineate the *Eco*RI fragments used in this experiment. Numbers are in base pairs. *c*, *d*, Autoradiographs of DBM paper-bound RNase H-resistant RNA fragments probed with 5′-end-specific (1.1 kilobase *Eco*RI fragment from pMSa104; *c*) or 3′-end-specific (4.4 kilobase *Eco*RI fragment from pMSa104; *d*) labelled DNA restriction fragments (see *b*). Poly(A)$^+$ RNA from pancreas (P, 500 ng), salivary gland (S, 1 μg) and liver (L, 40 μg) was hybridized to 100 ng (300 ng for pancreas) of the 520-base pair *Eco*RI restriction fragment from pMSa104 (*b*) in a final volume of 100 μl of 80% formamide, 0.05% SDS, 0.4 M NaCl, 1 mM EDTA, 10 mM PIPES, *p*H 6.4, for 3 h at 45 °C. After ethanol precipitation, the mRNA/DNA hybrids were digested with RNase H for 15 min at 37 °C in 10 mM Tris-HCl *p*H 7.6, 0.13 M NH_4Cl, 0.01 M Mg acetate, 5% (w/v) sucrose, 20 μg ml^{-1} oligo(dT), and 1 unit RNase H (Enzo) in a final volume of 30 μl (8 units RNase H in 300 μl for liver). After phenol extracting and ethanol precipitating, equivalent amounts of RNase H-resistant α-amylase mRNA sequences were electrophoresed on a 2% methyl mercury hydroxide agarose gel, transferred to DBM paper[10,20] and hybridized with the 5′- or 3′-end-specific probes. Numbers are in nucleotides, the 1,580-nucleotide bands in lane P represent uncleaved, deadenylated α-amylase mRNA. *e*, A 20-fold longer exposure of lane L in *d*. An arrow indicates the minor species fragment.

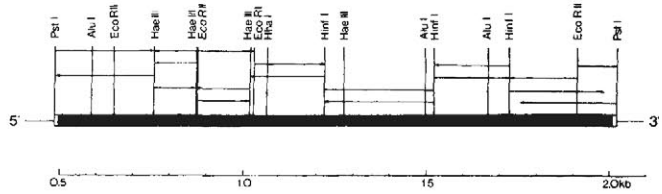


Fig. 2 Restriction map and strategy used to determine the nucleotide sequence of the liver α-amylase cDNA insert of recombinant plasmid pMLa52. Restriction endonuclease cleavage sites were determined by the technique of Smith and Birnstiel[21]. 5′-end-labelled fragments were sequenced according to Maxam and Gilbert[16]. The direction and extent of sequence information determined for each fragment examined are indicated by an arrow. The black box represents mRNA sequences, the open boxes indicate poly(dG-dC) generated during the cloning procedure. kb, kilobases.

Therefore, the entire sequence of the cDNA of pMLa52 was determined according to Maxam and Gilbert[16] as outlined in Fig. 2 legend. Nucleotides 518–1,773 of the liver α-amylase mRNA (Fig. 3) were elucidated from pMLa52. These residues are identical to those determined for the salivary gland α-amylase mRNA[11]. As 3′-minor nucleotide sequences have been reported elsewhere[12], only those for the 3′-major species are included in Fig. 3.

To obtain primary structure information for 5′-terminal sequences not included in the cDNA clone, we used reverse transcriptase-catalysed primer extension as described by Hagenbüchle *et al.*[11]. A 50-nucleotide single-stranded DNA fragment, isolated from the salivary gland α-amylase cDNA clone pMSa104 and containing nucleotide residues 191–241 (Fig. 3), was 5′-end-labelled according to Maxam and Gilbert[16] and hybridized to liver polyadenylated RNA. Two cDNA transcripts were produced after incubation with AMV reverse transcriptase which differed by approximately 30 nucleotides. The sizes and relative amounts of the two cDNAs were consistent with those predicted by the RNase H sizing experiment for the two liver mRNAs (Fig. 1). A unique sequence was elucidated for the 5′-major species cDNA (residues 1–191, Fig. 3) and simultaneous determination of the cytosine and purine residues of the 5′-minor species cDNA revealed that it is an extended version of the 5′-major species (data not shown). Notably, nucleotides 159–191 (Fig. 3) are identical to those determined for the comparable portion of the salivary gland α-amylase mRNA. Due to the low cellular concentration of liver α-amylase mRNA, direct RNA sequencing by chemical decapping and 5′-end labelling was not feasible; therefore, the 5′-terminal few residues could not be determined.

Sequences determined for the liver α-amylase cDNA produced by primer extension (nucleotides 1–191, Fig. 3) do not overlap those elucidated from pMLa52 DNA (nucleotides 518–1,773, Fig. 3). Of these, residues 159–191 and 518–1,773 are identical to those determined for the salivary gland α-amylase mRNA. We assume that the two α-amylase mRNAs also share residues 192–517 (sequences not examined for the liver species) because S_1 nuclease digestion and thermal melting curves of liver α-amylase mRNA/salivary gland α-amylase cDNA hybrids reveal no sequence heterogeneity[10].

Features of the liver α-amylase mRNAs

The results obtained by a combination of RNase H digestion, primed cDNA synthesis and sequence analysis indicate that several α-amylase mRNA species accumulate in the liver (Fig. 4). The 1,773-nucleotide major mRNA (5′-major/3′-major) accounts for 80% of the total α-amylase mRNA population. The remaining 20% are composed of 5′-minor species which contain ~30 additional nucleotides at their extreme 5′ termini. Among these two mRNA species, 5% contain the 237 3′-minor species residues; the distribution of 3′-minor species among 5′-major and 5′-minor mRNAs has not been determined. We do not know whether these minor species serve some special function or whether they represent products of alternative synthesis or processing pathways.

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L (m^7Gppp)XCCAUCAUAGGUCACUGUGGAGCUCAGAUCACAGUGCUGACAGAAUCCAUAUUUGGAGAAUUACAUAAGGUUUGAAAGAGAGGAUAGUGAAAGGAUACG 100

AAUUCCUAAAAACGUUUAAUCUGGCCUUUUGUUUGAACGAAAGAGAAAUUGAAACCAA
AUGAAAUAAAUUAGUUGUUAGAAAGAAUACUGCCAACAGCAU 200
S (m^7Gppp)AUUCUCAACGUAAAUCAGAAGAUUCCAACUGCAGUGGAAGGCAGCAC

AGCAAAAUGAAAUUCUUCCUGCUGCUUUCCCUCAUUGGAUUCUGCUGGGCCCAAUAUGACCCACAUACUCAAUAUGGACGAACUGCUAUUAUCCACCUGU 300
MetLysPhePheLeuLeuLeuSerLeuIluGlyPheCysTrpAlaGlnTyrAspProHisThrGlnTyrGlyArgThrAlaIluIluHisLeuP

UUGAGUGGCGCUGGGUUGAUAUUGCUAAGGAAUGUGAGAGAUACUUAGCUCCUAAUGGAUUUGCAGGUGUGCAGGUCUCUCCACCCAAUGAAAACAUCGU 400
heGluTrpArgTrpValAspIluAlaLysGluCysGluArgTyrLeuAlaProAsnGlyPheAlaGlyValGlnValSerProProAsnGluAsnIluVa

AGUCCACAGCCCUUCAAGACCAUGGUGGGAAAGAUAUCAACCAAUUAGCUACAAAAUAUGUUCCAGGUCUGGAAAUGAAGAUGAAUUCAGGGACAUGGUG 500
lValHisSerProSerArgProTrpTrpGluArgTyrGlnProIluSerTyrLysIluCysSerArgSerGlyAsnGluAspGluPheArgAspMetVal

AACAGGUGCAACAAUGUUGGUGUCCGUAUUUAUGUGGAUGCUGUCAUUAACCACAUGUGUGGAGUGGGGGCUCAAGCUGGACAAAGCAGUACAUGUGGAA 600
AsnArgCysAsnAsnValGlyValArgIluTyrValAspAlaValIluAsnHisMetCysGlyValGlyAlaGlnAlaGlyGlnSerSerThrCysGlyS

GUUAUUUCAACCCAAAUAACAGGGACUUUCCUGGAGUUCCCUAUUCUGGUUUUGACUUUAAUGAUGGAAAAUGUAGAACUGCAAGUGGAGGUAUCGAGAA 700
erTyrPheAsnProAsnAsnArgAspPheProGlyValProTyrSerGlyPheAspPheAsnAspGlyLysCysArgThrAlaSerGlyGlyIluGluAs

CUACCAAGAUGCUGCUCAGGUCAGAGAUUGUCGUCUGUCUGGCCUUCUGGAUCUUGCACUUGAGAAAGAUUAUGUUCGAACCAAGGUGGCUGACUAUAUG 800
nTyrGlnAspAlaAlaGlnValArgAspCysArgLeuSerGlyLeuLeuAspLeuAlaLeuGluLysAspTyrValArgThrLysValAlaAspTyrMet

AACCAUCUCAUUGACAUUGGCGUAGCAGGGUUCAGACUUGAUGCUUCUAAGCACAUGUGGCCUGGAGACAUAAAGGCAAUUUUGGACAAACUGCAUAAUC 900
AsnHisLeuIluAspIluGlyValAlaGlyPheArgLeuAspAlaSerLysHisMetTrpProGlyAspIluLysAlaIluLeuAspLysLeuHisAsnL

UCAAUACAAAAUGGUUCUCCCAAGGAAGCAGACCUUUCAUUUUCCAAGAGGUGAUUGAUCUGGGUGGUGAGGCAGUGUCAAGUAAUGAGUAUUUUGGAAA 1,000
ε nThrLysTrpPheSerGlnGlySerArgProPheIluPheGlnGluValIluAspLeuGlyGlyGluAlaValSerSerAsnGluTyrPheGlyAs

UGGCCGUGUGACAGAAUUCAAAUAUGGAGCAAAAUUGGGCAAAGUUAUGCGCAAGUGGGAUGGAGAAAAGAUGUCCUACUUAAAGAACUGGGGAGAAGGU 1,100
nGlyArgValThrGluPheLysTyrGlyAlaLysLeuGlyLysValMetArgLysTrpAspGlyGluLysMetSerTyrLeuLysAsnTrpGlyGluGly

UGGGGUUUGAUGCCUUCUGACAGAGCCCUUGUGUUUGUGGACAACCAUGACAAUCAGCGAGGACAUGGUGCUGGGGGAGCAUCCAUCUUGACAUUCUGGG 1,200
TrpGlyLeuMetProSerAspArgAlaLeuValPheValAspAsnHisAspAsnGlnArgGlyHisGlyAlaGlyGlyAlaSerIluLeuThrPheTrpA

AUGCUAGACUCUAUAAAAUGGCUGUUGGCUUUAUGUUGGCUCAUCCUUAUGGUUUCACACGGGUGAUGUCAAGUUACUAUUGGCCAAGAAAUUUCCAGAA 1,300
spAlaArgLeuTyrLysMetAlaValGlyPheMetLeuAlaHisProTyrGlyPheThrArgValMetSerSerTyrTyrTrpProArgAsnPheGlnAs

UGGAAAAGAUGUCAAUGACUGGGUUGGACCACCAAAUAACAAUGGAAAAACCAAAGAAGUGAGCAUUAACCCAGACAGCACUUGUGGCAAUGACUGGAUC 1,400
nGlyLysAspValAsnAspTrpValGlyProProAsnAsnAsnGlyLysThrLysGluValSerIluAsnProAspSerThrCysGlyAsnAspTrpIlu

UGUGAACAUCGAUGGCGUCAAAUAAGGAACAUGGUUGCCUUCAGAAAUGUCGUCAAUGGUCAGCCUUUUGCAAACUGGUGGGAUAAUGACAGCAACCAGG 1,500
CysGluHisArgTrpArgGlnIluArgAsnMetValAlaPheArgAsnValValAsnGlyGlnProPheAlaAsnTrpTrpAspAsnAspSerAsnGlnV

UAGCUUUUGGCAGAGGAAACAAAGGACUCAUUGUCUUUAACAAUGAUGACUGGGCUUUGUCAGAAACUUUACAGACUGGUCUUCCUGCUGGCACAUACUG 1,600
alAlaPheGlyArgGlyAsnLysGlyLeuIluValPheAsnAsnAspAspTrpAlaLeuSerGluThrLeuGlnThrGlyLeuProAlaGlyThrTyrCy

UGAUGUCAUUUCUGGAGAUAAAGUCGAUGGCAAUUGCACUGGAAUAAAAGUCUAUGUUGGCAAUGAUGGCAAAGCUCACUUUUCUAUUAGUAACUCUGCC 1,700
sAspValIluSerGlyAspLysValAspGlyAsnCysThrGlyIluLysValTyrValGlyAsnAspGlyLysAlaHisPheSerIluSerAsnSerAla

GAAGACCCAUUUAUUGCAAUCCAUGCAGAGUCAAAAAUAUAAAUUUAAAAUAAACACAUAUUGAGAGCAUCA$_n$
GluAspProPheIluAlaIluHisAlaGluSerLysIlu

Fig. 3 Comparison of the nucleotide sequence of the major liver α-amylase mRNA and its salivary gland counterpart. Numbering begins with nucleotide X (which represents the few initial nucleotides that were not determined, see text) in the liver mRNA. The nucleotide residues 112–158 shown for the salivary gland mRNA were taken from O.H. *et al.*[11] but contain two nucleotides (residues 113 and 114) that were excluded from the sequence reported earlier. The two mRNAs are assumed to be capped. S and L indicate the tissue-specific salivary gland and 5′-major liver α-amylase mRNA leaders. The first AUG codon is underlined; that actually used to initiate α-amylase synthesis is boxed, as is the terminator triplet. Both mRNAs predict the amino acid sequence shown below the nucleotide sequence.

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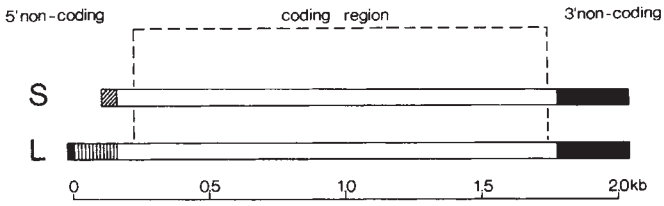

Fig. 4 Schematic comparison of the liver and salivary gland α-amylase mRNAs. The diagram is based on data presented here and elsewhere[11,12], as described in the text. The open boxes represent common sequences; the black boxes designate residues found in the minor salivary (S) and liver (L) α-amylase mRNA species. Tissue-specific leader residues are shaded diagonally for salivary gland mRNA or vertically for the liver mRNA species. Numbering is that in Fig. 3.

Tissue-specific 5′-noncoding segments

Figure 3 compares the salivary gland α-amylase mRNA sequence with that of its major liver counterpart. The two mRNAs share nucleotides 159–1,773, which include the entire coding and 3′-noncoding regions. These salivary gland mRNA sequences have been discussed in detail previously[11]. Briefly, translation of the α-amylase precursor can occur from only one coding frame (the two others contain many termination triplets); this frame predicts a precursor protein whose characteristics mirror those determined for α-amylase produced *in vivo* and *in vitro*[10,17].

In addition to the coding and 3′-noncoding regions, a portion of the 5′-non-translated region is identical in the liver and salivary gland α-amylase mRNAs. The 204- and 93-nucleotide 5′-terminal noncoding regions share the 48 residues immediately adjacent the coding region. This common segment (nucleotides 159–206, Fig. 3) has the intriguing feature that it begins with an AUG codon which is not used to initiate translation of α-amylase even though it is the one nearest the 5′ terminus. The first AUG is followed by a lysine codon, then the termination triplet UAA. The AUG used to initiate translation[11] lies 14 codons downstream. As all other eukaryotic cellular mRNAs for which complete sequence information is available initiate translation at the AUG codon located nearest the 5′ end[18], the salivary gland and liver α-amylase mRNAs are apparent exceptions. Whether the first AUG in these α-amylase mRNAs has some regulatory role in translation is unclear.

It is the initial 158 nucleotides of the major liver species and the 47 5′-terminal residues of its salivary gland counterpart that distinguish the α-amylase mRNAs which accumulate in these two tissues. Hence, these two species have distinct 5′-terminal noncoding regions but direct the translation of an identical α-amylase polypeptide. The presence of distinguishing 5′-non-translated sequences might be the consequence of tissue-specific requirements for the rate of translation initiation. Although the influence of particular sequences 5′ to the AUG initiation codon is unclear[18], translation initiation efficiency *in vitro* can be affected by the length of the 5′-non-translated region (C. Queen, personal communication). Therefore, the different lengths of the 5′-non-translated segments in salivary gland and liver α-amylase mRNAs (93 and 204 residues, respectively) might influence their relative translatability. Finally, the distinct 5′-non-translated segments might also specify different lifetimes for these tissue-specific mRNAs.

The observation that liver and salivary gland α-amylase mRNAs share identical coding and 3′-noncoding residues suggests that they are transcribed from the same DNA sequence. Indeed, recent analysis of genomic DNA demonstrates that both liver and salivary gland α-amylase mRNAs are specified by a single DNA sequence in the mouse haploid genome[19]. Two different segments of DNA specify the distinct 5′-non-translated regions of these mRNAs. These findings have implications for the mechanisms which govern the expression of this α-amylase gene; for example, tissue-specific splice events must be responsible for the correct biosynthesis of α-amylase mRNA in the liver and salivary gland.

We thank B. Hirt for his support, S. Cherpillod for typing the manuscript, D. Petrelli for technical assistance; L. J. Korn and B. Meyer for comments on the manuscript, and J. Beard for supplying reverse transcriptase through the Office of Program Resources and Logistics, Viral Cancer Program (Viral Oncology Division of Cancer Cause and Prevention), NCI. This work was supported by grant 3.148.77 from the Swiss NSF.

Received 29 October 1980; accepted 13 January 1981.

1. Loyter, A. & Schramm, M. *Biochim. biophys. Acta* **65**, 200–206 (1962).
2. Arnold, M. & Rutter, W. J. *J. biol. Chem.* **238**, 2760–2765 (1963).
3. Hjorth, J. P., Meisler, M. H. & Nielsen, J. T. *Genetics* **92**, 915–930 (1979).
4. Messer, M. & Dean, R. I. *Biochem. J.* **151**, 17–22 (1975).
5. Sick, K. & Nielsen, J. T. *Hereditas* **51**, 291–296 (1964).
6. Hammerton, K. & Messer, M. *Biochim. biophys. Acta* **244**, 441–451 (1971).
7. Sanders, T. G. & Rutter, W. J. *Biochemistry* **11**, 130–136 (1972).
8. Kaplan, R. D., Chapman, V. & Ruddle, F. H. *J. Hered.* **64**, 155–157 (1973).
9. Takeuchi, T., Matsushima, T. & Sugimura, T. *Biochim. biophys. Acta* **403**, 122–130 (1975).
10. Schibler, U., Tosi, M., Pittet, A.-C. & Wellauer, P. K. *J. molec. Biol.* **142**, 93–116 (1980).
11. Hagenbüchle, O., Bovey, R. & Young, R. A. *Cell* **21**, 179–187 (1980).
12. Tosi, M., Young, R. A., Hagenbüchle, O. & Schibler, U. *Nucleic Acids Res.* (submitted).
13. Donis-Keller, H. *Nucleic Acids Res.* **7**, 179–192 (1979).
14. Robertson, H. D. & Dunn, J. J. *J. biol. Chem.* **250**, 3030–3056 (1975).
15. Sippel, A. E., Stavrianopoulos, J. G., Schütz, G. & Feigelson, P. *Proc. natn. Acad. Sci. U.S.A.* **71**, 4635–4639 (1974).
16. Maxam, A. M. & Gilbert, W. *Meth. Enzym.* **65**, 499–560 (1980).
17. Danielsson, A., Marklund, S. & Stigbrand, T. *Int. J. Biochem.* **6**, 585–589 (1975).
18. Kozak, M. *Cell* **15**, 1109–1123 (1978).
19. Young, R. A., Hagenbüchle, O. & Schibler, U. Cell (in the press).
20. Alwine, J. C., Kemp, D. J. & Stark, G. R. *Proc. natn. Acad. Sci. U.S.A.* **74**, 5350–5354 (1977).
21. Smith, H. O. & Birnstiel, M. L. *Nucleic Acids Res.* **3**, 2387–2398 (1976).

Control of vascular permeability by polymorphonuclear leukocytes in inflammation

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Polymorphonuclear leukocytes infiltrate tissues in response to an inflammatory stimulus to remove invading micro-organisms and cell debris. We present evidence that these scavenging cells have another, more sophisticated role in that they are involved in the control of fluid efflux through the blood vessel wall which leads to tissue oedema.

SINCE the first observations of leukocytes accumulating at a site of inflammation, their relationship to changes occurring in blood vessels has been a source of controversy. Studies of animal models of inflammation (for example, rat paw oedema in response to carrageenin[1]) have suggested that acute vascular responses, vasodilatation and increased vascular permeability, result from the sequential release of low molecular weight mediators—histamine, serotonin, bradykinin and prostaglandins[2]. During these acute vascular changes polymorphonuclear (PMN) leukocytes, predominantly neutrophils, accumulate

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0028–0836/81/080646—05$01.00

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slowly in the tissues reaching significant numbers after several hours. Based on these models, and allergic models such as the Arthus reaction[3], several mechanisms have been proposed whereby PMN leukocytes could contribute to oedema. The most widely accepted of these, first postulated by Metchnikoff[4], was that during phagocytosis PMN leukocytes release substances (lysosomal enzymes[5] for example) which damage vascular endothelial cells—the cells lining the blood vessel wall which are normally responsible for retaining plasma proteins within the lumen. We suggest an alternative hypothesis, that PMN leukocytes, responding to a chemical signal in the tissue, can control the permeability of the blood vessel wall to plasma proteins. Three such signalling substances are described here. Although PMN leukocytes are present in high numbers in tissues several hours after the initiation of an inflammatory response, the evidence suggests that these cells interact with vascular endothelial cells to regulate permeability within a few minutes of the inflammatory stimulus.

The chemical mediators of inflammatory oedema in man are unknown; there is little evidence that histamine, serotonin and bradykinin are important. We suggest that the mechanisms involving leukocytes described here may be more relevant.

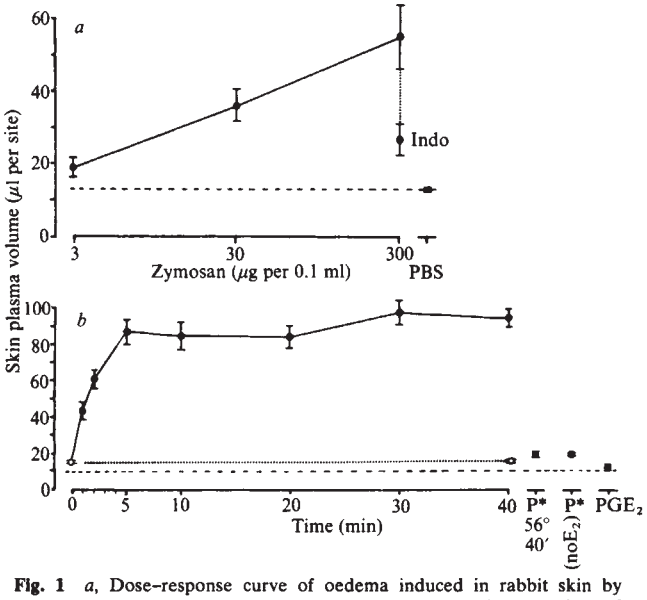

Fig. 1 *a*, Dose-response curve of oedema induced in rabbit skin by intradermal injection of zymosan. *b*, Time course of generation of permeability-increasing activity in blood plasma incubated with zymosan at 37°C. In all experiments plasma exudation was measured using the accumulation of intravenously injected ^{131}I-human serum albumin in rabbit (male NZW, 3–3.5 kg) dorsal skin in response to 0.1-ml intradermal injections of the agents. After a fixed period (30 min in all experiments except where stated) animals were killed, the skin was removed, the injection sites were excised (16-mm diameter punch) and counted in a γ-counter. *a* Shows the increase in exudate volume (measured over a 90-min period) in response to increasing amounts of injected zymosan. Indomethacin (Indo), an inhibitor of prostaglandin synthesis, injected locally at 0 min and 60 min (3×10^{-9} mol per site) suppressed exudation, as shown , an effect which was reversed by exogenous prostaglandin[14]. Oedema induced by intradermally injected zymosan was also suppressed by nitrogen mustard (1.75 mg per kg intravenous injection, 4 days before testing in the skin) which depletes circulating PMN leukocytes. Plasma volumes (90 min accumulation) of skin sites injected with 100 μg of zymosan were 40.0 ± 6.3 μl in control animals, and 13.1 ± 2.1 μl in depleted animals ($n=4$ rabbits, saline control (PBS, phosphate-buffered saline) values 16.6 ± 1.8 μl and 15.7 ± 2.0 μl, respectively). Unlike suppression of oedema produced by indomethacin, the effect of nitrogen mustard was not reversed by exogenous prostaglandin. In *b*, following incubation of plasma with zymosan for periods up to 40 min, zymosan was removed by centrifugation and the plasma was mixed with PGE_2 (3×10^{-10} mol per site) before injection into the skin. Exudation was measured over a 30-min period. Permeability-increasing activity was rapidly generated in plasma during incubation (response $t_{1/2}=100$ s) but oedema was only demonstrable when PGE_2 was present. Little exudation was induced by the following: plasma incubated alone, tested with PGE_2 (open symbols); plasma incubated with zymosan for 40 min but tested without PGE_2 (P* no E_2); or PGE_2 alone. Preheating plasma to 56 °C for 40 min (P* 56°40′) before the addition of zymosan prevented the generation of activity (40 min incubation with zymosan and the plasma tested with PGE_2). All the results are shown as the mean ±s.e.m. for six replicate injections. Values for PBS controls are shown as dashed lines (as in all the figures).

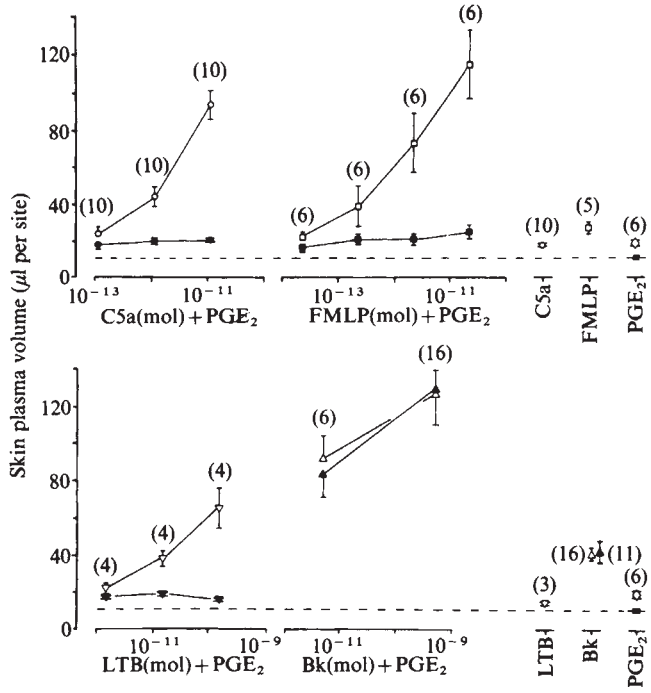

Fig. 2 Oedema induced in rabbit skin by rabbit C5a, FMLP, leukotriene B_4 (LTB) and bradykinin (Bk); the potentiating effect of PGE_2; and the inhibitory effect of leukocyte depletion. All responses were measured over a 30-min period. Numbers in parentheses refer to the number of animals used in each test; the mean response to six replicate injections of each substance was determined for each animal. Open symbols show responses in untreated animals. Closed symbols show responses in animals treated 4 days previously with a single intravenous injection of nitrogen mustard (1.75 mg per kg). The symbols on the right-hand side show responses to C5a, FMLP, leukotriene B_4 and bradykinin at the highest doses used in the experiments. PGE_2 was used at a dose of 3×10^{-10} mol per site throughout. In normal rabbits (PMN leukocyte count $3.5\pm1.1\times10^6$ per ml blood, $n=6$ rabbits) very small responses were elicited by C5a, FMLP, leukotriene B_4 and PGE_2 when tested alone (20–30 μl responses to LTB_4 alone at higher doses have been reported[30]). However, when mixed with PGE_2 the leukotactic substances were very potent in inducing oedema. Bradykinin produced distinct responses alone and these were enhanced by PGE_2. In PMN leukocyte-depleted rabbits (PMN leukocyte count $4.0\pm1.4\times10^4$ per ml blood, $n=6$ rabbits), although responses to bradykinin and bradykinin+PGE_2 were unaffected, responses to the leukotactic substances in combination with PGE_2 were virtually abolished. Responses to histamine+PGE_2, like those to bradykinin+PGE_2, were unaffected by PMN-leukocyte depletion—histamine (2.3×10^{-8} mol per site)+PGE_2 (3×10^{-10} mol per site) produced a response of 82.7 ± 2.8 μl in controls and 78.0 ± 14.5 μl in treated animals ($n=4$ rabbits).

Prostaglandins and oedema formation

The presence of microorganisms in a tissue stimulates leakage of plasma from small blood vessels resulting in tissue oedema. We investigated the mechanisms underlying this vascular response by injecting yeast cell walls (zymosan) into rabbit dorsal skin and measuring plasma leakage using the local accumulation of intravenously injected ^{131}I-albumin[6]. Zymosan induces acute oedema with a peak rate of plasma exudation at 1–3 h, decreasing to low levels by 4–5 h (ref. 7). Figure 1*a* shows that the volume of oedema fluid accumulating is regulated according to the stimulus size (the quantity of zymosan injected).

We deduced that zymosan stimulates the release in the skin of two chemicals which exert a synergistic action to mediate oedema ('the two mediator hypothesis'[8]). The first mediator increases vascular permeability by an action involving vascular endothelial cells; the second greatly potentiates plasma exudation by inducing relaxation of arteriolar smooth muscle cells and thus increasing the blood supply to the tissue. The vasodilator mediator is thought to be a prostaglandin[7–10] (PGE_2 or PGI_2)[9] produced by cell membranes; little exudation is induced when prostaglandin synthesis is inhibited (for example, by indomethacin[11]) as shown in Fig. 1*a*.

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Involvement of the complement system

We next investigated the identity of the endogenous permeability-increasing mediator. We obtained evidence[12-14] that this mediator is derived from the complement system—the enzyme system in blood plasma and tissue fluid which is responsible for lysis of microorganisms. As shown in Fig. 1*b*, incubation of blood plasma with zymosan at 37°C resulted in the rapid generation of a substance which could increase vascular permeability when injected into skin. This substance induced little oedema unless mixed with a vasodilator prostaglandin. The active substance has been purified and identified[14] as a polypeptide fragment of the fifth component of complement, C5a. The activity is stable in plasma and is therefore probably due to C5a devoid of the carboxyl-terminal arginine (C5a des Arg); the arginine group, which is necessary for histamine-releasing (anaphylatoxic) activity in other species, is cleaved by plasma carboxypeptidase B (ref. 15). We have shown that oedema induced by synergism between rabbit C5a and prostaglandin is not dependent on histamine release[12,14].

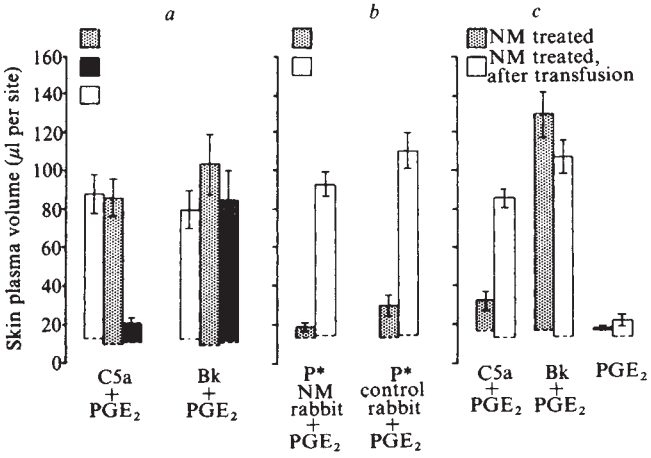

Fig. 3 *a*, Results of an experiment designed to show that nitrogen mustard suppresses responses in skin by an action on dividing bone marrow cells, rather than by an action on the skin itself. Rabbits were set up in three groups of four individuals. The first group were untreated (open columns). The second group were injected, via an ear vein, with nitrogen mustard (1.75 mg per kg) while a clamp was applied to the terminal descending aorta (stippled columns) and the clamp was removed after 10 min. As nitrogen mustard has a very short half life in the circulation, this procedure allowed access of nitrogen mustard to the dorsal skin but not to the bone marrow of the hind limbs. The third group were sham-operated and injected with nitrogen mustard via an ear vein (hatched columns). As in the second group a volume of blood (17% of total blood volume, measured by isotope dilution) was not exposed to nitrogen mustard; this volume of blood was withdrawn from the individuals of the sham-operated group before drug injection, maintained at 37 °C and reinjected after 10 min. Four days later, agents were injected into the skin and responses measured over a 30-min period. All three groups responded to bradykinin (5×10^{-10} mol per site) + PGE_2 (3×10^{-10} mol per site). Animals whose skin had been exposed to nitrogen mustard but whose bone marrow had been protected (group 2) responded normally to C5a (1.5×10^{-11} mol per site) + PGE_2 (3×10^{-10} mol per site) showing that inhibition of responses by nitrogen mustard was not due to an effect of the drug on the skin. *b*, Results of an experiment designed to investigate the effect of nitrogen mustard (NM) on C5a generation in plasma. Rabbits were set up in two groups of five individuals, one group injected with nitrogen mustard (stippled), the other untreated (open columns). Four days later plasma from each animal was incubated with zymosan (1 mg ml^{-1}, 37 °C for 30 min). The zymosan was then removed by centrifugation. Plasma was mixed with PGE_2 (3×10^{-10} mol per site) and responses measured in the skin (30 min period) of the donor animal and in an animal in the other group. Treated animals did not respond to plasma from either group; however, untreated animals responded to plasma from both groups. These results demonstrate that nitrogen mustard pretreatment suppresses responses to C5a, but not its production. *c*, Results of an experiment to show skin responses in a depleted animal before, and after, cross blood transfusion with a normal animal. Six such experiments were carried out; three depleted animals showed only a small elevation in circulating PMN leukocytes following transfusion and these animals did not respond to C5a + PGE_2. In three other experiments the circulating PMN leukocyte count was elevated following transfusion and these recipient animals responded to C5a + PGE_2. ^{125}I-albumin was injected intravenously into both normal and depleted animals. C5a (1.5×10^{-11} mol per site) + PGE_2 (3×10^{-10} mol per site) and bradykinin (5×10^{-10} mol per site) + PGE_2 (3×10^{-10} mol per site) were then injected intradermally into the depleted animal. After 30 min, blood was transfused (6 ml min^{-1}) by a peristaltic pump interposed between carotid artery cannulae and ear vein cannulae. After a further 60 min, pumping was stopped, the treated animal was injected intravenously with ^{131}I-albumin, and the skin was again tested with the same combinations of agents. Following transfusion the circulating PMN leukocyte count was elevated from 3×10^4 ml^{-1} to 2.4×10^6 ml^{-1}; only a small change was seen in the mononuclear leukocyte count, from 2.3×10^6 to 3.2×10^6 ml^{-1}. The animal clearly responded to C5a + PGE_2 following transfusion. Results shown as mean ± s.e.m. for six replicate injections. Hatched bars, nitrogen mustard-treated; open bars, nitrogen mustard-treated after transfusion.

Central role for the PMN leukocyte

In both intact and des Arg forms, C5a shows powerful chemoattraction for PMN leukocytes *in vitro*[16]. Thus, is the permeability-increasing activity of C5a in the skin dependent on the presence of PMN leukocytes *in vivo*? Zymosan failed to induce oedema in the skin of rabbits depleted of circulating PMN leukocytes using nitrogen mustard (results in Fig. 1 legend), providing the first indication that the action of C5a might be leukocyte dependent.

To investigate this further, the activity of histamine and bradykinin (both with established permeability-increasing activity) was compared with that of three leukotactic substances[16-18] in the skin of normal animals and animals depleted of circulating PMN leukocytes. The leukotactic substances used were: rabbit C5a (ref. 14), a polypeptide of approximately 70 amino acids; leukotriene B_4, a dihydroxy fatty acid formed from arachidonic acid by the action of leukocyte lipoxygenase[19]; and *N*-formyl-methionyl-leucyl-phenylalanine (FMLP), a synthetic substance based on formyl peptides secreted by bacteria in culture[18]. Figure 2 shows that in normal animals leukotactic substances alone induced little oedema, but, mixed with PGE_2, all three were highly potent permeability-increasing substances. However, in animals given intravenous nitrogen mustard 4 days before testing, to deplete circulating PMN leukocytes[20,21], the leukotactic substances were ineffective, even when combined with PGE_2. In normal animals both bradykinin and histamine induced some oedema when injected alone into the skin and the responses were greatly enhanced by PGE_2. However, these responses were unaffected by nitrogen mustard pretreatment. These results suggested that bradykinin and histamine increase vascular permeability by acting directly on vascular endothelial cells ('direct-action mediators'), but that C5a, leukotriene B_4 and FMLP can only act in the presence of PMN leukocytes ('polymorph-dependent mediators').

Nitrogen mustard (administered as a single intravenous injection) is short-lived in the blood and depletes circulating PMN leukocytes by preventing replication of stem cells in bone marrow. However, as the skin itself is exposed to the drug, a direct effect on the skin could not be excluded. A clamp applied to the terminal descending aorta during, and for 10 min after, an ear-vein injection of nitrogen mustard provided access for the drug to the skin, but not to the bone marrow of the hind limbs. As Fig. 3*a* shows, 4 days later animals treated in this way responded normally to C5a + PGE_2. Thus, suppression of responses by nitrogen mustard was not due to an effect on the skin. Furthermore, 4 days after nitrogen mustard treatment, the numbers of circulating mononuclear cells and platelets were little affected[20] and, as shown in Fig. 3*b*, the plasma of animals pretreated with nitrogen mustard could be activated normally to produce permeability-increasing activity, that is, C5a (complement levels measured by haemolytic assay are also known to be unaffected[20]).

All these results suggested that PMN leukocytes are essential for the permeability response to the leukotactic substances. Nevertheless, we could not raise circulating cell numbers sufficiently to restore responsiveness in previously depleted

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animals by intravenous or intra-arterial injections of PMN leukocytes. This probably indicates the importance of the surface properties of the cells which may change following removal from the body. Experiments using whole blood were more successful: in three out of six experiments it was possible to elevate circulating levels of PMN leukocytes in depleted rabbits by whole-blood cross-transfusion (6 ml min^{-1} for 60 min) using normal donor rabbits. Figure 3*c* shows that responsiveness to C5a was restored in these rabbits.

In all experiments Evans blue dye was mixed with ^{131}I-albumin to indicate plasma leakage sites in the skin. Responses to different agents were visually similar; however, direct-action mediators could be clearly distinguished from polymorph-dependent mediators by their time courses of action using radioisotope techniques, as shown in Fig. 4. Oedema was first detectable at 1.5 min after injection of bradykinin + PGE_2 but at 6 min with C5a + PGE_2 (surprisingly early considering the necessity for PMN leukocytes). A more striking difference was that responses to bradykinin and histamine were of very short duration, as expected from earlier experiments[22] (for example, bradykinin $t_{1/2} = 4$ min), whereas those to leukotactic substances were protracted (for example, C5a $t_{1/2} = 95$ min).

From our results we conclude that mechanisms exist which regulate local fluid accumulation in response to the presence of microorganisms in tissue. This process, although remarkably fast in onset, seems to be dependent on PMN leukocytes. PMN leukocytes are attracted to the site and their function is controlled by chemicals released in the tissue. The chemical signals can be generated in response to microorganisms in tissue fluid (for example, C5a), can be derived from microorganisms themselves (for example, FMLP), or, perhaps in later phases of the response, derived from leukocytes (for example, leukotriene B_4). These substances induce little oedema alone; the concomitant generation of vasodilator prostaglandin seems to be essential for oedema formation.

How do PMN leukocytes increase vascular permeability?

The manner in which PMN leukocytes increase vessel wall permeability remains undetermined. PMN leukocytes, during their passage through the vessel wall, may secrete enzymes which break down substances present at junctions between adjacent endothelial cells, or the enzymes may act on basement membrane underlying endothelial cells. Alternatively, PMN leukocytes may secrete a substance which affects endothelial cells, perhaps causing contraction. If PMN leukocytes do secrete such a substance, phagocytosis does not seem to be essential because soluble factors (C5a, FMLP and leukotriene B_4) or particulate matter (zymosan) can induce increased vascular permeability equally well.

Secretion by PMN leukocytes may be unnecessary as electron micrographs of tissues removed shortly after injection of C5a showed PMN leukocytes between the endothelial cell layer and basement membrane, many underneath endothelial cell junctions. Direct contact of that sort may be required for endothelial cells to respond. If PMN leukocytes located under junctions were to swell (as has been observed in response to C5a *in vitro*[23]) this could open the junctions.

PMN leukocytes in other types of inflammatory reaction?

We believe that these mechanisms have developed as an essential part of the host defence system to control the supply of complement from the blood to an infected tissue in order to regulate local killing of microorganisms. Similar mechanisms probably operate in allergic inflammation to regulate the supply of complement, antibodies and other plasma components to the tissue. In many models of inflammation oedema is known to be suppressed by prostaglandin synthesis inhibitors, complement depletion and PMN leukocyte depletion; these observations are consistent with our hypothesis.

Pulmonary oedema in man is often associated with a local accumulation of PMN leukocytes, for example in adult respiratory distress syndrome due to septicaemia, during haemodialysis and cardiopulmonary bypass[24]. There is evidence that the interaction of C5a with PMN leukocytes is important in these conditions[24,25].

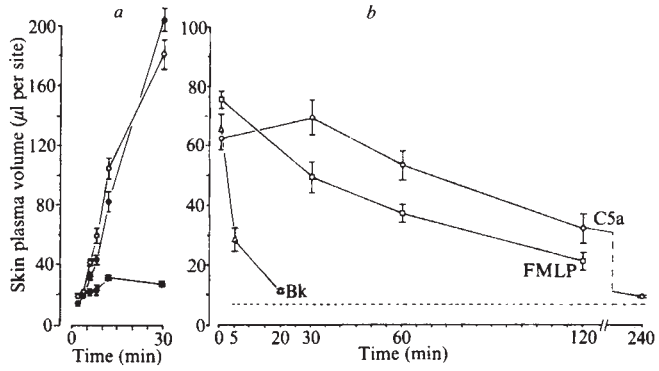

Fig. 4 The onset time of the vascular permeability increase induced by injection of C5a (*a*), and the duration of increased permeability induced by C5a and FMLP (*b*). *a*, Rabbits were injected intravenously with ^{131}I-albumin and killed 30 min later. Intradermal injections were given at intervals (shown in the abscissa) before death. The injections given were: ○, C5a (1.5×10^{-11} mol per site) + PGE_2 (3×10^{-10} mol per site); ●, C5a + PGE_2 + mepyramine maleate (3.5×10^{-9} mol per site); and ◆, PGE_2 alone. Six minutes after injection, responses to C5a + PGE_2 were significantly greater than control values produced by PGE_2 alone ($P < 0.01$, Student's unpaired *t*-test). Mepyramine, at doses which abolished matching responses to histamine + PGE_2, had a limited suppressor effect on responses; responses to C5a + PGE_2 + mepyramine were also significantly elevated above controls at 6 min ($P < 0.01$). Bradykinin and histamine responses were earlier in onset, for example bradykinin (5×10^{-10} mol per site) + PGE_2 (3×10^{-10} mol per site) onset time 1.5 min ($P < 0.01$). In the experiments shown in *b*, bradykinin (5×10^{-10} mol per site), C5a (1.5×10^{-11} mol per site) and FMLP (2.3×10^{-11} mol per site) were injected at intervals (as shown in abscissa) before a local superimposed injection of PGE_2 (3×10^{-10} mol per site). Labelled albumin was injected intravenously immediately before the PGE_2 injection and responses measured over a period of 30 min. (This procedure is designed to negate the effect of PGE_2 metabolism on the result.) Clearly the durations of action of the leukotactic substances were quite different from that of bradykinin: bradykinin $t_{1/2} = 4$ min, C5a $t_{1/2} = 95$ min, and FMLP $t_{1/2} = 50$ min. Histamine responses were similar in duration to those of bradykinin (results not shown). Results are represented as mean ± s.e.m., $n = 4$ replicates in *a*, and $n = 6$ replicates in *b*.

In chronic, self-destructive inflammation (such as rheumatoid arthritis), mononuclear cells are intimately involved with the underlying pathogenesis, and these cells could interact with the vessel wall in the manner we have described. In such chronic conditions, PMN leukocytes may be important because phases of active oedema formation are associated with the appearance of high numbers of these cells in the tissues[26]. Our simple model may be relevant to these complex events as rheumatoid fluid has been shown to contain prostaglandins[27], leukotriene B_4 (ref. 28) and C5a (ref. 29).

In summary, we suggest that PMN leukocytes and endothelial cells lining the blood vessel wall cooperate to regulate the supply of macromolecules to a tissue, and that this may represent a fundamental component of the inflammatory response.

We thank our colleagues in the Department of Pharmacology, Royal College of Surgeons, for discussions. C5a, from zymosan-activated rabbit plasma, was purified and characterized in collaboration with Dr P. J. Jose[14]. We thank Dr A. W. Ford-

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Hutchinson and his colleagues for the gift of leukotriene B_4, and M. J. Peck and M. J. Forrest for technical assistance. This work was supported by the MRC.

Received 12 August; accepted 2 December 1980.

Note added in proof: The synergistic effect of leukotriene B_4 and PGE_2 has been confirmed by G. A. Higgs (personal communication) and in several species by M. A. Bray *et al.*[31].

1. Winter, C. A., Risley, E. A. & Nuss, G. V. *Proc. Soc. exp. Biol. Med.* **111**, 544 (1962).
2. Di Rosa, M., Giroud, J. P. & Willoughby, D. A. *J. Path. Bact.* **104**, 15–29 (1971).
3. Arthus, M. *C.r. Séanc. Soc. Biol.* **55**, 817 (1903).
4. Metchnikoff, E. *Annls Inst. Pasteur, Paris* **1**, 321 (1887).
5. Movat, H. Z. *Exp. molec. Path.* **31**, 201–210 (1979).
6. Williams, T. J. *J. Physiol., Lond.* **254**, 4-5P (1976).
7. Williams, T. J. *Br. J. Pharmac.* **65**, 517–524 (1979).
8. Williams, T. J. & Peck, M. J. *Nature* **270**, 530–532 (1977).
9. Peck, M. J. & Williams, T. J. *Br. J. Pharmac.* **62**, 464–465P (1978).
10. Williams, T. J. & Morley, J. *Nature* **246**, 215–218 (1973).
11. Vane, J. R. *Nature new Biol.* **231**, 232–235 (1971).
12. Williams, T. J. *J. Physiol., Lond.* **281**, 44–45P (1978).
13. Jose, P. J., Peck, M. J., Robinson, C. & Williams, T. J. *J. Physiol., Lond.* **281**, 13–14P (1978).
14. Williams, T. J. & Jose, P. J. *J. exp. Med.* **153**, 136–153 (1981).
15. Hugli, T. E. & Müller-Eberhard, H. J. *Adv. Immun.* **26**, 1–53 (1978).
16. Fernandez, H. N., Henson, P. M., Otani, A. & Hugli, T. E. *J. Immun.* **120**, 109–115 (1978).
17. Ford-Hutchinson, A. W., Bray, M. A., Doig, M. V., Shipley, M. E. & Smith, M. J. H. *Nature* **286**, 264–265 (1980).
18. Schiffmann, E., Corcoran, B. A. & Wahl, S. M. *Proc. natn. Acad. Sci. U.S.A.* **72**, 1059–1062 (1975).
19. Samuelsson, B., Borgeat, P., Hammarström, S. & Murphy, R. C. *Prostaglandins* **17**, 785–787 (1979).
20. Humphrey, J. H. *Br. J. exp. Path.* **36**, 268–282 (1955).
21. Stetson, C.A. *J. exp. Med.* **94**, 347–348 (1951).
22. Williams, T. J. & Morley, J. *Br. J. exp. Path.* **55**, 1–12 (1974).
23. O'Flaherty, J. T., Kreutzer, D. L. & Ward, P. A. *Am. J. Path.* **90**, 537–550 (1978).
24. Larsen, G. L., McCarthy, K., Webster, R. O., Henson, J. & Henson, P. M. *Am. J. Path.* **100**, 179–192 (1980).
25. Hammerschmidt, D. E., Weaver, L. J., Hudson, L. D., Craddock, P. R. & Jacob, H. S. *Lancet* **i**, 947–949 (1980).
26. Ward, P. A. & Zvaifler, N. J. *J. clin. Invest.* **50**, 606–616 (1971).
27. Higgs, G. A., Vane, J. R., Hart, F. D. & Wotjulewski, J. A. in *Prostaglandin Synthetase Inhibitors* (eds Robinson, H. J. & Vane, J. R.) 165–174 (Raven, New York, 1974).
28. Klickstein, L. B., Shapleigh, C. & Goetzl, E. J. *J. clin. Invest.* **66**, 1166–1170 (1980).
29. Zvaifler, N. J. *J. exp. Med.* **134**, 276–285 (1971).
30. Eakins, K. E., Higgs, G. A., Moncada, S., Salmon, J. A. & Spayne, J.A. *J. Physiol., Lond.* **307**, 71P (1980).
31. Bray, M. A., Cunningham, F. M., Ford-Hutchinson, A. W. & Smith, M. J. H. *Br. J. Pharmac.* (in the press).

Nucleotide and corresponding amino acid sequences encoded by α and β tubulin mRNAs

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Most of the mRNA sequences coding for α and β tubulin in embryonic chick brain have been determined by sequencing of cloned cDNA copies of these mRNAs. From a 1,682-base pair cDNA sequence we have deduced the entire protein sequence for β tubulin. For α tubulin, all but about 38 N-terminal amino acids have been deduced from the cDNA sequence. Although tyrosine has previously been shown to be post-translationally added to the C-terminus of α tubulin by a specific ligase, we conclude that the primary post-translational event must be the removal, not the addition, of tyrosine because a terminal tyrosine is encoded by the mRNA.

TUBULIN is the principle subunit of microtubules, an intracellular cylindrical filamentous structure present in almost all eukaryotic cells. Microtubules function as structural and motile elements in mitosis, intracellular transport, flagellar movement and the cytoskeleton (for review see refs 1, 2). The microtubule, 24 nm in diameter, is comprised of 13 protofilaments each containing an alternating array of α and β tubulin subunits spaced every 4 nm. In the soluble form it is generally assumed that tubulin is a heterodimer composed of one α and one β chain each of molecular weight (MW) about 55,000.

The structure of the microtubule is highly conserved and at the resolution obtained by electron microscopy (~2 nm) no differences in structure have been observed between microtubules from *Tetrahymena*, sea urchin and mammalian sources. The tubulin subunit will form copolymers across wide species lines and the conditions for assembly of tubulin are similar for each species. Recently, we have shown that cDNA clones for chicken embryonic α and β tubulin (which do not cross-hybridize to each other) will hybridize in stringent conditions to genomic DNA from sea urchin and *Drosophila*, as well as mammals[3,4], and in less stringent conditions to yeast, *Chlamydomonas* and trypanosomes (N. Neff, D. Botstein, D. Cleveland and N. Agabian, unpublished results).

This high degree of conservation is not surprising in view of the large number of conserved structural interactions of tubulin. The tubulin dimer has two GTP binding sites, one exchangeable and the other not, a single colchicine binding site, two binding sites for vinblastine, and other drug binding sites. In addition, the tubulin structure must be constrained by the requirement for interaction with other tubulin molecules in the microtubule lattice as well as with microtubule-associated proteins.

Despite considerable recent interest in the biochemistry of tubulin, there are important gaps in our knowledge of its primary structure and genetic complexity. Evidence suggesting some homology and possible common evolutionary origin of the α and β peptides relies on the published N-terminal 25 amino acids in α and β tubulin[5]. Additional other known data are the sequence of some unordered peptides[6] and the 78 C-terminal amino acids of the α chain[7]. A true appreciation of the degree of homology between α and β chains, however, awaits more complete sequence information. Additional sequencing studies are also crucial to understanding the microheterogeneity, post-translational modification and structure of the tubulin proteins themselves.

Recently, we have constructed cDNA clones from embryonic mRNA of chicken brain and have isolated clones containing α or β tubulin sequences. We report here the complete DNA sequence of the cDNA from these clones. The β tubulin clone

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contains the entire coding sequence for the protein and extensive 5′ and 3′ untranslated regions of the mRNA. The α clone contains >90% of the coding sequences and some of the 3′ untranslated region of the α tubulin mRNA.

DNA sequences of plasmids pT2 and pT1

The construction of bacterial plasmids containing sequences homologous to α or β tubulin mRNAs from embryonic chick brain has been described elsewhere[3]. We initially demonstrated by mRNA hybridization followed by *in vitro* translation of specifically bound mRNA that the hybrid plasmids pT2 and pT1 carry β and α tubulin sequences, respectively. The sequencing strategies for both pT1 and pT2 are presented in Fig. 1. All sequencing was done by the method of Maxam and Gilbert[8]. In all cases both strands of the cDNAs were sequenced unless a particular single-strand sequence was exceptionally clear.

The sequence of the 1,681 nucleotides of the cDNA from pT2, the β tubulin plasmid, is given in Fig. 2*a*, along with the corresponding protein sequence which it predicts. The direction and frame of reading were established by comparison with the published N-terminal 25-amino acid sequence for chick brain β tubulin[5] and by the presence of the 3′ poly(A) tract at one end of the cDNA. In addition, the C-terminus of the protein was established by the appearance of three in-phase stop codons and by the presence of a C-terminal peptide with a composition (15 out of 25 residues acidic) similar to that reported for α tubulin[7]. Plasmid pT2 thus contains the complete coding sequence for the 445 amino acids of β tubulin as well as 87 nucleotides 5′ to the methionine initiation codon and 226 nucleotides between the UGA stop codon and the site of polyadenylation. The predicted MW of β tubulin is 49,935 and the sequence deduced agrees with 19 of the 25 N-terminal residues reported by Luduena and Woodward[5] for chick brain β tubulin. The cDNA sequence predicts that residues 11 and 15 are glutamic acid, whereas glutamine was the amino acid residue previously reported[5] at these positions. Similarly, we predict that residues 10, 13 and 17 are glycine whereas threonine was originally reported at these positions.

The sequence of the chick brain α tubulin clone, pT1, is given in Fig. 2*b*. The direction and frame of reading were established by identification of the known C-terminal protein sequences for porcine brain α tubulin. Although pT1 contains 123 nucleotides 3′ to the UGA stop codon, no poly(A) tract is present; therefore some regions of the 3′ untranslated region of the mRNA may be missing. Furthermore, pT1 does not contain the complete 5′

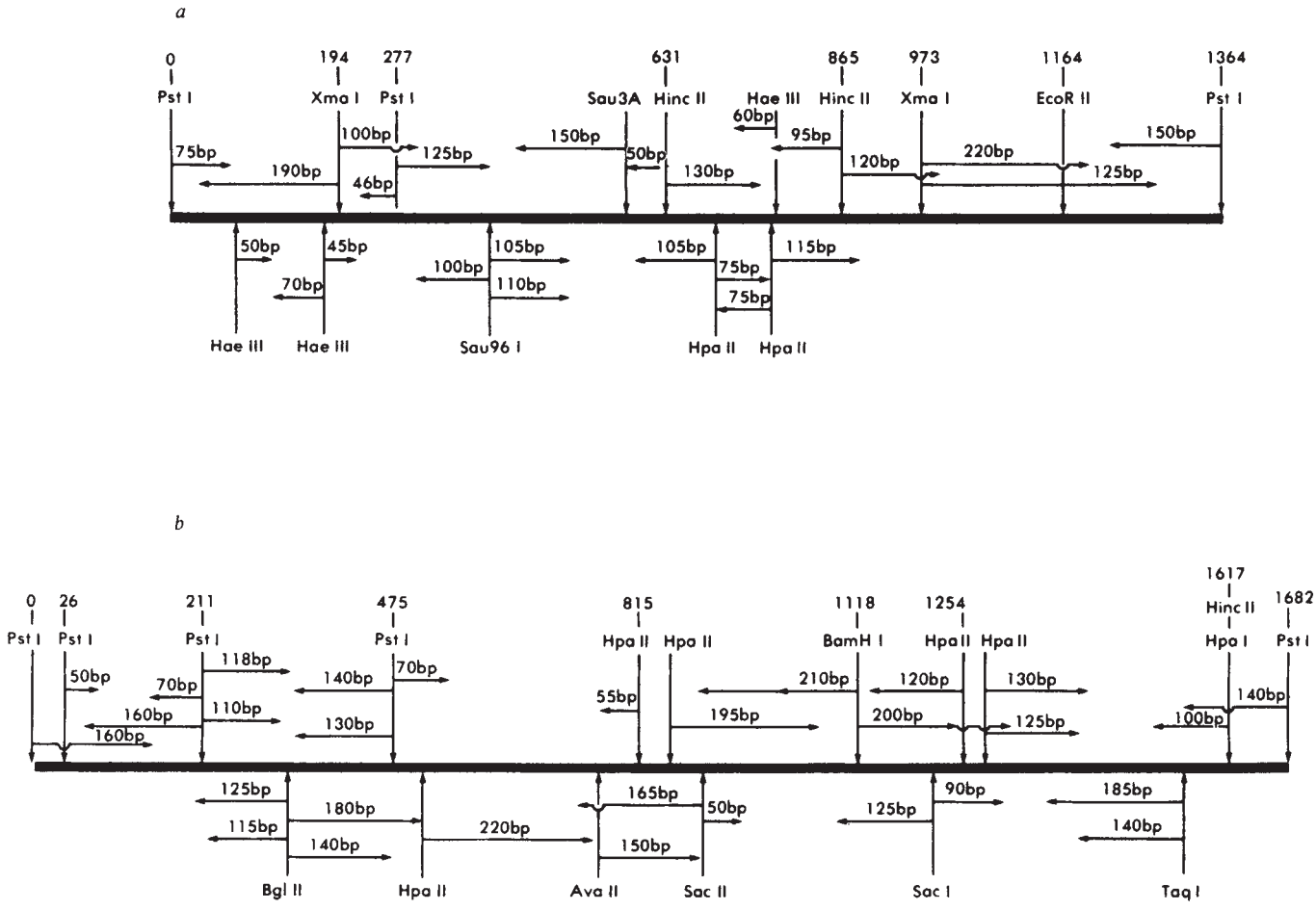

Fig. 1 Sequencing strategy for the chicken brain α and β tubulin cDNAs. *a*, α tubulin cDNA; *b*, β tubulin cDNA. Nucleotide residues are numbered in the direction from 5′ to 3′ in the message strand. The extent and direction of each fragment sequenced is shown by the horizontal arrows. Only the restriction sites used as starting points are shown. For end labelling, DNA was treated with 5 μg of bacterial alkaline phosphatase (Worthington) at 37 °C for 60 min in 20 mM Tris-HCl, *p*H 8.0, phenol-extracted and treated with 10 units of T4 polynucleotide kinase (Boehringer) at 37 °C for 30 min in a reaction mixture containing 50 mM glycine, *p*H 9.5, 10 mM $MgCl_2$, 10 mM dithiothreitol, 0.1 mM spermidine and 0.001 mM [γ-^{32}P]ATP (~3,500 Ci $mmol^{-1}$). Fragments labelled at only one end were isolated by gel electrophoresis after either digestion with a restriction enzyme or strand separation. DNA sequencing was carried out by the method of Maxam and Gilbert[8].

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a UAAGGCCCGGAGCGGGGCGCGGCCGCUGCAGCGGAGAGCGGGAGGUGACGGAGCGGGAGCAGCGCGGCACCGGCAGACACCGGCAUC AUG CGU GAG AUC GUG CAC AUC CAG

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    10   20   30
Ala Gly Gln Cys Gly Asn Gln Ile Gly Ala Lys Phe Trp Glu Val Ile Ser Asp Glu His Gly Ile Asp Pro Thr Gly Ser Tyr His Gly
GCC GGC CAG UGC GGC AAC CAG AUC GGC GCU AAG UUC UGG GAG GUC AUC AGC GAU GAG CAC GGC AUC GAU CCC ACC GGC AGC UAC CAC GGG

    40   50   60
Asp Ser Asp Leu Gln Leu Glu Arg Ile Asn Val Tyr Tyr Asn Glu Ala Thr Gly Asn Lys Tyr Val Pro Arg Ala Ile Leu Val Asp Leu
GAC AGC GAC CUG CAG CUG GAG AGG AUC AAC GUG UAC UAC AAU GAG GCC ACA GGU AAC AAG UAC GUC CCC CGU GCC AUC CUG GUU GAC CUG

    70   80   90
Glu Pro Gly Thr Met Asp Ser Val Arg Ser Gly Pro Phe Gly Gln Ile Phe Arg Pro Asp Asn Phe Val Phe Gly Gln Ser Gly Ala Gly
GAG CCC GGC ACG AUG GAC UCG GUG CGC UCC GGC CCC UUU GGA CAG AUC UUC CGA CCC GAC AAC UUU GUC UUU GGU CAG AGC GGG GCC GGC

    100   110   120
Asn Asn Trp Ala Lys Gly His Tyr Thr Glu Gly Ala Glu Leu Val Asp Ser Val Leu Asp Val Val Arg Lys Glu Ser Glu Ser Cys Asp
AAC AAC UGG GCC AAG GGG CAC UAC ACG GAA GGC GCU GAG CUG GUG GAC UCU GUG CUG GAU GUG GUG AGG AAG GAG UCG GAA AGC UGC GAC

    130   140   150
Cys Leu Gln Gly Phe Gln Leu Thr His Ser Leu Gly Gly Gly Thr Gly Ser Gly Met Gly Thr Leu Leu Ile Ser Lys Ile Arg Glu Glu
UGC CUG CAG GGC UUC CAG UUG ACC CAC UCG CUG GGC GGC GGC ACC GGC UCG GGG AUG GGC ACC CUC CUC AUC AGC AAG AUC CGC GAG GAG

    160   170   180
Tyr Pro Asp Arg Ile Met Asn Thr Phe Ser Val Met Pro Ser Pro Lys Val Ser Asp Thr Val Val Glu Pro Tyr Asn Ala Thr Leu Ser
UAC CCC GAC CGC AUC AUG AAC ACG UUC AGC GUC AUG CCC UCC CCC AAG GUG UCG GAC ACG GUG GUG GAG CCC UAC AAU GCC ACC CUU UCU

    190   200   210
Val His Gln Leu Val Glu Asn Thr Asp Glu Thr Tyr Cys Ile Asp Asn Glu Ala Leu Tyr Asp Ile Cys Phe Arg Thr Leu Lys Leu Thr
GUG CAC CAG CUG GUG GAG AAC ACG GAC GAG ACC UAC UGC AUC GAC AAC GAG GCC CUG UAC GAC AUC UGC UUC CGC ACC CUG AAG CUC ACC

    220   230   240
Thr Pro Thr Tyr Gly Asp Leu Asn His Leu Val Ser Ala Thr Met Ser Gly Val Thr Thr Cys Leu Arg Phe Pro Gly Gln Leu Asn Ala
ACU CCC ACG UAC GGG GAC CUC AAC CAC CUG GUG UCG GCC ACC AUG AGC GGC GUG ACC ACC UGC CUU CGC UUC CCC GGC CAG CUG AAC GCC

    250   260   270
Asp Leu Arg Lys Leu Ala Val Asn Met Val Pro Phe Pro Arg Leu His Phe Phe Met Pro Gly Phe Ala Pro Leu Thr Ser Arg Gly Ser
GAC CUG CGC AAG CUG GCG GUC AAC AUG GUG CCU UUC CCC CGG CUG CAC UUC UUC AUG CCG GGC UUC GCC CCG CUG ACG AGC CGC GGC AGC

    280   290   300
Gln Gln Tyr Arg Ala Leu Thr Val Pro Glu Leu Thr Gln Gln Met Phe Asp Ser Lys Asn Met Met Ala Ala Cys Asp Pro Arg His Gly
CAG CAG UAC CGA GCC CUG ACG GUG CCC GAG CUG ACG CAG CAG AUG UUC GAC UCC AAG AAC AUG AUG GCC GCC UGC GAC CCC CGC CAC GGC

    310   320   330
Arg Tyr Leu Thr Val Ala Ala Ile Phe Arg Gly Arg Met Ser Met Lys Glu Val Asp Glu Gln Met Leu Asn Val Gln Asn Lys Asn Ser
CGC TAC CUG ACG GUG GCU GCC AUC UUC CGA GGC CGC AUG UCC AUG AAG GAG GUG GAC GAG CAG AUG CUG AAC GUG CAG AAC AAG AAC AGC

    340   350   360
Ser Tyr Phe Val Glu Trp Ile Pro Asn Asn Val Lys Thr Ala Val Cys Asp Ile Pro Pro Arg Gly Leu Lys Met Ser Ala Thr Phe Ile
AGC UAC UUU GUG GAG UGG AUC CCC AAC AAC GUG AAG ACG GCC GUC UGC GAC AUC CCC CCG CGU GGC CUC AAG AUG UCC GCC ACC UUC AUC

    370   380   390
Gly Asn Ser Thr Ala Ile Gln Glu Leu Phe Lys Arg Ile Ser Glu Gln Phe Thr Ala Met Phe Arg Arg Lys Ala Phe Leu His Trp Tyr
GGC AAC AGC ACG GCC AUC CAG GAG CUC UUC AAG AGG AUC UCG GAG CAG UUC ACG GCC AUG UUC CGG CGC AAG GCC UUC UUG CAC UGG UAC

    400   410   420
Thr Gly Glu Gly Met Asp Glu Met Glu Phe Thr Glu Ala Glu Ser Asn Met Asn Asp Leu Val Ser Glu Tyr Gln Gln Tyr Gln Asp Ala
ACC GGC GAG GGC AUG GAU GAG AUG GAG UUC ACC GAG GCG GAG AGC AAC AUG AAC GAC CUG GUC UCA GAG UAC CAG CAA UAC CAG GAU GCC

    430   440   445
Thr Ala Asp Glu Gln Gly Glu Phe Glu Glu Glu Gly Glu Glu Asp Glu Ala OP
ACC GCU GAU GAG CAG GGU GAA UUU GAA GAG GAA GGA GAG GAG GAU GAG GCA UGA AGUGGAAAUGGGUAGGAGUCAAGUGUAGUCUGAGCAGGCAAGUGUUCA
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UUGAGAGGAGAAUCUGUGCAGUUGUGCUGAAGCAUGCGUUUCUUAAUUUGGUGCUCUCCGCUGUUGCUUUUGUCAGCAGUUUUGUAUCCUCGACUGUCCGAUGUAACAGUUGCAAAAAUA

CUUCAGAGUCUUCUGUUGAAAUGGUUAACUUCUCAAACAUAAAGGCUUUUUGUGUUCUAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAA

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b
    40                                                  50                                                  60
Thr Ile Gly Gly Gly Asp Asp Ser Phe Asn Thr Phe Phe Ser Glu Thr Gly Ala Gly Lys His Val Pro Arg Ala Val Phe Val Asp
GAA ACC AUC GGC GGG GAC GAC UCC UUC AAC ACC UUC UUC AGC GAG ACG GGG GCC GGC AAG CAC GUG CCC CGU GCC GUC UUU GUG GAC

    70                                                  80                                                  90
Leu Glu Pro Thr Val Ile Asp Glu Val Arg Thr Gly Thr Tyr Arg Gln Leu Phe His Pro Glu Gln Leu Ile Thr Gly Lys Glu Asp Ala
CUG GAG CCC ACC GUG AUC GAU GAG GUG CGC ACG GGG ACG UAC CGG CAG CUC UUC CAC CCC GAG CAG CUG AUC ACG GGC AAG GAG GAU GCG

    100                                                 110                                                 120
Ala Asn Asn Tyr Ala Arg Gly His Tyr Thr Ile Gly Lys Glu Ile Ile Asp Leu Val Leu Asp Arg Ile Arg Lys Leu Ala Asp Gln Cys
GCC AAC AAC UAC GCC CGG GGG CAC UAC ACC AUC GGC AAG GAG AUC AUC GAC CUG GUG CUG GAC CGC AUC CGC AAG CUG GCU GAC CAG UGC

    130                                                 140                                                 150
Thr Gly Leu Gln Gly Phe Ser Val Phe His Ser Phe Gly Gly Gly Thr Gly Ser Gly Phe Thr Ser Leu Leu Met Glu Arg Leu Ser Val
ACG GGG CUG CAG GGC UUC UCG GUG UUC CAC AGC UUC GGG GGC GGC ACC GGC UCC GGC UUC ACC UCG CUG CUG AUG GAG CGC CUG UCG GUG

    160                                                 170                                                 180
Asp Tyr Gly Lys Lys Ser Lys Leu Glu Phe Ser Ile Tyr Pro Ala Arg Gln Val Ser Thr Ala Val Val Glu Pro Tyr Asn Ser Ile Leu
GAC UAC GGC AAG AAG UCC AAG CUG GAG UUC UCC AUC UAC CCG GCC CGU CAG GUG UCC ACG GCC GUG GUG GAG CCC UAC AAC UCC AUC CUC

    190                                                 200                                                 210
Thr Thr His Thr Thr Leu Glu His Ser Asp Cys Ala Phe Met Val Asp Asn Glu Ala Ile Tyr Asp Ile Cys Arg Arg Asn Leu Asp Ile
ACC ACC CAC ACC ACG CUG GAG CAC UCC GAC UGC GCC UUC AUG GUG GAC AAC GAG GCC AUC UAC GAC AUC UGU CGC AGG AAC CUG GAC AUC

    220                                                 230                                                 240
Glu Arg Pro Thr Tyr Thr Asn Leu Asn Arg Leu Ile Gly Gln Ile Val Ser Ser Ile Thr Ala Ser Leu Arg Phe Asp Gly Ala Leu Asn
GAG CGC CCC ACC UAC ACC AAC CUC AAC AGG CUG AUA GGG CAG AUC GUG UCC UCC AUC ACA GCC UCA CUG CGC UUC GAU GGG GCU CUG AAC

    250                                                 260                                                 270
Val Asp Leu Thr Glu Phe Gln Thr Asn Leu Val Pro Tyr Pro Arg Ile His Phe Pro Leu Ala Thr Tyr Ala Pro Val Ile Ser Ala Glu
GUC GAC CUG ACG GAG UUC CAG ACC AAC CUG GUG CCC UAC CCU CGC AUC CAC UUC CCA UUG GCC ACG UAU GCG CCG GUG AUC UCU GCU GAG

    280                                                 290                                                 300
Lys Ala Tyr His Glu Gln Leu Ser Val Ala Glu Ile Thr Asn Ala Tyr Phe Glu Pro Ala Asn Gln Met Val Lys Cys Asp Pro Arg His
AAG GCC UAC CAC GAG CAG CUG UCG GUG GCU GAG AUC ACC AAC GCG UAC UUC GAG CCG GCC AAC CAG AUG GUG AAG UGC GAC CCG CGG CAC

    310                                                 320                                                 330
Gly Lys Tyr Met Ala Cys Cys Leu Leu Tyr Arg Gly Asp Val Val Pro Lys Asp Val Asn Ala Ala Ile Ala Thr Ile Lys Thr Lys Arg
GGC AAG UAC AUG GCG UGC UGC CUG CUG UAC CGC GGG GAC GUG GUG CCC AAA GAU GUC AAC GCC GCC AUC GCC ACC AUC AAG ACC AAG CGC

    340                                                 350                                                 360
Thr Ile Gln Phe Val Asp Trp Cys Pro Thr Gly Phe Lys Val Gly Ile Asn Tyr Gln Pro Pro Thr Val Val Pro Gly Gly Asp Leu Ala
ACC AUU CAG UUC GUA GAC UGG UGC CCA ACU GGU UUC AAG GUG GGC AUC AAC UAC CAG CCU CCC ACC GUG GUG CCC GGG GGG GAC CUG GCC

    370                                                 380                                                 390
Lys Val Gln Arg Ala Val Cys Met Leu Ser Asn Thr Thr Ala Ile Ala Glu Ala Trp Ala Arg Leu Asp His Lys Phe Asp Leu Met Tyr
AAG GUG CAG CGC GCC GUG UGC AUG CUG AGC AAC ACC ACG GCC AUC GCC GAG GCG UGG GCG CGC UUG GAC CAC AAG UUU GAC CUG AUG UAC

    400                                                 410                                                 420
Ala Lys Arg Ala Phe Val His Trp Tyr Val Gly Glu Gly Met Glu Glu Gly Glu Phe Ser Glu Ala Arg Glu Asp Met Ala Ala Leu Glu
GCC AAG CGC GCC UUC GUG CAC UGG UAU GUG GGG GAG GGC AUG GAG GAG GGG GAG UUC UCA GAG GCU CGG GAG GAU AUG GCU GCC CUG GAG

    430                                                 440                                                 450
Lys Asp Tyr Glu Glu Val Gly Val Asp Ser Val Glu Gly Glu Gly Glu Glu Glu Gly Glu Glu Tyr OP
AAG GAU UAC GAG GAG GUG GGG GUG GAU UCC GUG GAG GGU GAG GGU GAG GAG GAA GGA GAG GAG UAC UGA GAGUUCACAACCGUCCUGCUCGUACAGGGA
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ACAAUCCUAGUUAUCCCCCACCCCACCCCCGUGGGUCUGUAGCGGUGUGCGCUGCUGUCUGUGCUGUUCUGUGGUCUGUGGUUCAAUGGAAAGUC

Fig. 2 *a*, The nucleotide sequence of chicken brain β tubulin mRNA, determined from the double-stranded cDNA insert of pT2 (ref. 3). The predicted complete amino acid sequence is shown above the RNA sequence. *b*, The partial nucleotide sequence of chicken brain α tubulin mRNA, determined from the double-stranded cDNA insert of pT1 (ref. 3). The predicted partial amino acid sequence of α tubulin is also indicated. The amino acids have been numbered by comparison with the complete sequence of β tubulin mRNA shown in *a*.

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sequences of the mRNA. By alignment with the deduced β tubulin protein sequence from above, we predict that pT1 lacks the sequences coding for amino acids 1–38 of α tubulin. We used this assumption tentatively to number the amino acids for the α chain in Fig. 2*b*. The predicted MW for α tubulin is ~50,040. The predicted sequence corresponds to the existing protein sequence data in the following respects: (1) with the exception of the terminal tyrosine residue (see below), the C-terminal 78 amino acids reported for porcine brain α tubulin are identical to the chick sequence reported here, and (2) the sequences of four (α4D, α4HB, α4I and α4CA) of the eight unordered cyanogen bromide fragments reported for beef brain α tubulin[6] are identical to peptides in the chick sequence. One of the remaining reported peptides, α4J, contains a single substitution at position 302 in our sequence, and one peptide (α4HA) is actually a β tubulin peptide (residues 148–164).

The tubulin subunits have been widely seen to migrate on SDS or SDS-urea polyacrylamide gels with mobilities characteristic of proteins of MW ~55,000–56,000 for α tubulin and 53,000–55,000 for β tubulin (for example, see ref. 9). Comparable mobilities have been found for the *in vitro* translation products

a	2/UUU/Phe	1/UCU/Ser	2/UAU/Tyr	1/UGU/Cys
	18/UUC/Phe	9/UCC/Ser	17/UAC/Tyr	7/UGC/Cys
	0/UUA/Leu	2/UCA/Ser	0/UAA/OC	1/UGA/OP
	2/UUG/Leu	5/UCG/Ser	0/UAG/AM	3/UGG/Trp
	0/CUU/Leu	2/CCU/Pro	0/CAU/His	2/CGU/Arg
	3/CUC/Leu	9/CCC/Pro	11/CAC/His	13/CGC/Arg
	0/CUA/Leu	2/CCA/Pro	0/CAA/Gln	0/CGA/Arg
	24/CUG/Leu	4/CCG/Pro	12/CAG/Gln	4/CGG/Arg
	1/AUU/Ile	1/ACU/Thr	0/AAU/Asn	0/AGU/Ser
	21/AUC/Ile	18/ACC/Thr	15/AAC/Asn	3/AGC/Ser
	1/AUA/Ile	1/ACA/Thr	1/AAA/Lys	0/AGA/Arg
	8/AUG/Met	10/ACG/Thr	17/AAG/Lys	2/AGG/Arg
	0/GUU/Val	6/GCU/Ala	7/GAU/Asp	3/GGU/Gly
	3/GUC/Val	22/GCC/Ala	18/GAC/Asp	13/GGC/Gly
	1/GUA/Val	0/GCA/Ala	1/GAA/Glu	1/GGA/Gly
	28/GUG/Val	6/GCG/Ala	33/GAG/Glu	15/GGG/Gly

b	5/UUU/Phe	2/UCU/Ser	0/UAU/Tyr	0/UGU/Cys
	18/UUC/Phe	5/UCC/Ser	16/UAC/Tyr	8/UGC/Cys
	0/UUA/Leu	1/UCA/Ser	0/UAA/OC	1/UGA/OP
	2/UUG/Leu	7/UCG/Ser	0/UAG/AM	4/UGG/trp
	2/CUU/Leu	1/CCU/Pro	0/CAU/His	3/CGU/Arg
	6/CUC/Leu	16/CCC/Pro	10/CAC/His	11/CGC/Arg
	0/CUA/Leu	0/CCA/Pro	1/CAA/Gln	3/CGA/Arg
	22/CUG/Leu	3/CCG/Pro	21/CAG/Gln	2/CGG/Arg
	0/AUU/Ile	1/ACU/Thr	2/AAU/Asn	0/AGU/Ser
	19/AUC/Ile	15/ACC/Thr	21/AAC/Asn	14/AGC/Ser
	0/AUA/Ile	1/ACA/Thr	0/AAA/Lys	0/AGA/Arg
	19/AUG/Met	13/ACG/Thr	15/AAG/Lys	3/AGG/Arg
	1/GUU/Val	4/GCU/Ala	7/GAU/Asp	3/GGU/Gly
	7/GUC/Val	20/GCC/Ala	19/GAC/Asp	25/GGC/Gly
	0/GUA/Val	1/GCA/Ala	5/GAA/Glu	2/GGA/Gly
	21/GUG/Val	2/GCG/Ala	31/GAG/Glu	5/GGG/Gly

Fig. 4 Codon usage in α and β tubulin mRNAs. The numbers indicate the frequency with which individual codons are used in the coding regions of the mRNAs. OC, OP and AM designate the terminator codons ochre, opal and amber, respectively. *a*, α tubulin mRNA; *b*, β tubulin mRNA.

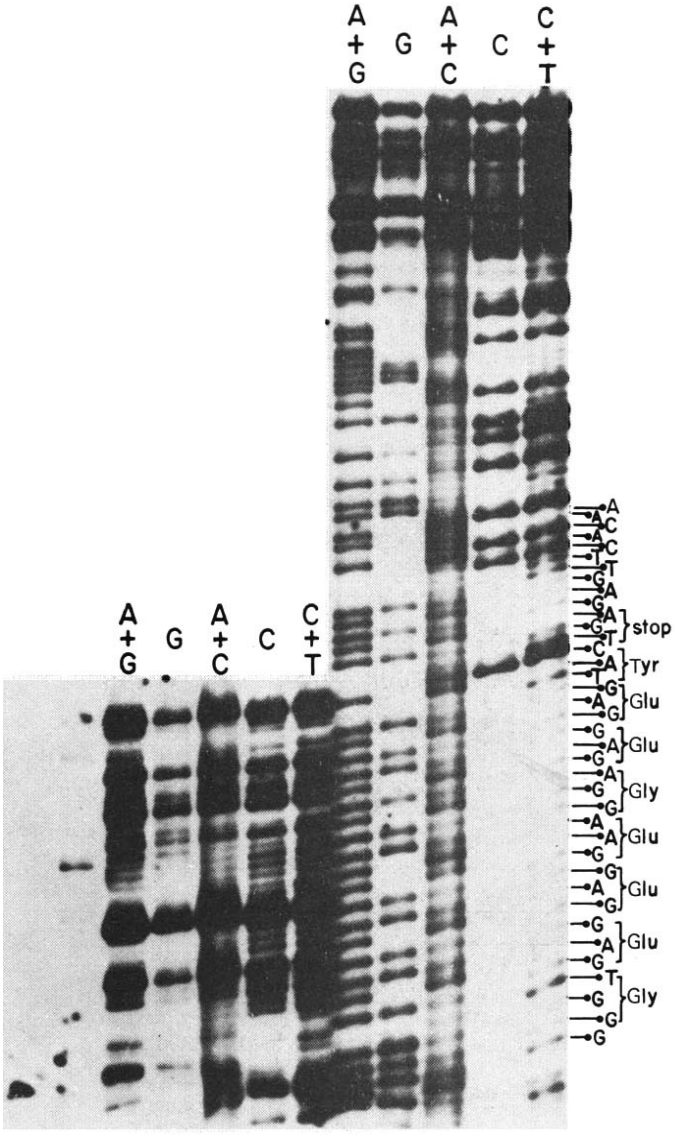

Fig. 3 Autoradiogram of a sequencing gel showing the region coding for the C-terminus of α tubulin. Note that the last codon before the UGA stop codon is UAC, which codes for tyrosine.

of α and β tubulin from mRNA translated in reticulocyte or wheat-germ systems (for example, see ref. 10). Surprisingly, we have unambiguously determined that the polypeptide encoded by one β tubulin mRNA has a MW of 49,935. Similarly, although we cannot be completely certain of the size of the α tubulin chain, by homology with the β tubulin sequence and by comparison with unpublished protein sequence data of porcine brain α tubulin (H. Ponstingl and collaborators, unpublished results), we predict that the MW of the α tubulin chain is slightly greater than 50,000. Because the mobilities of *in vitro* synthesized chains are indistinguishable from those of the mature polypeptides, post-translational modification cannot account for the differences in estimated and actual molecular weights.

Note also that the β chain contains 19 methionines and the α chain probably contains 11 methionines (using the data of Luduena and Woodward[5] for amino acids 1–25 coupled with the sequence of cyanogen bromide fragment α4CB of Lu and Elzinga[6], which seems to be derived from amino acids 22–36 of the α chain of beef α tubulin). In agreement with previous results[5], this predicts a ratio in methionine contents of 1.7:1, that is, the expected ratio of incorporation of label into each

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chain if each are translated at comparable rates or in comparable amounts. This could be a useful number for investigators using methionine-labelled materials.

The C-terminal residue of α tubulin is tyrosine

An enzyme has recently been reported which is capable of post-translationally adding a tyrosine residue to the C-terminal end of α tubulin[11–14]. The enzyme is specific for α tubulin, requires ATP and functions in the presence of protein synthesis inhibitors. Purified α tubulin has been reported to contain between 15 and 50% tyrosine on the C-terminal end[7,8,15]. Historically, then, C-terminal tyrosylation has been viewed as a post-translational modification of the α chain. However, it is now clear from the nucleotide sequence that α tubulin mRNA contains a coded terminal tyrosine (Fig. 2*b*). The sequencing gel demonstrating the tyrosine UAC codon immediately before the UGA stop codon is shown in Fig. 3. Although the functional significance of tyrosylation and detyrosylation remains unclear, these new data demonstrate that the primary post-translational

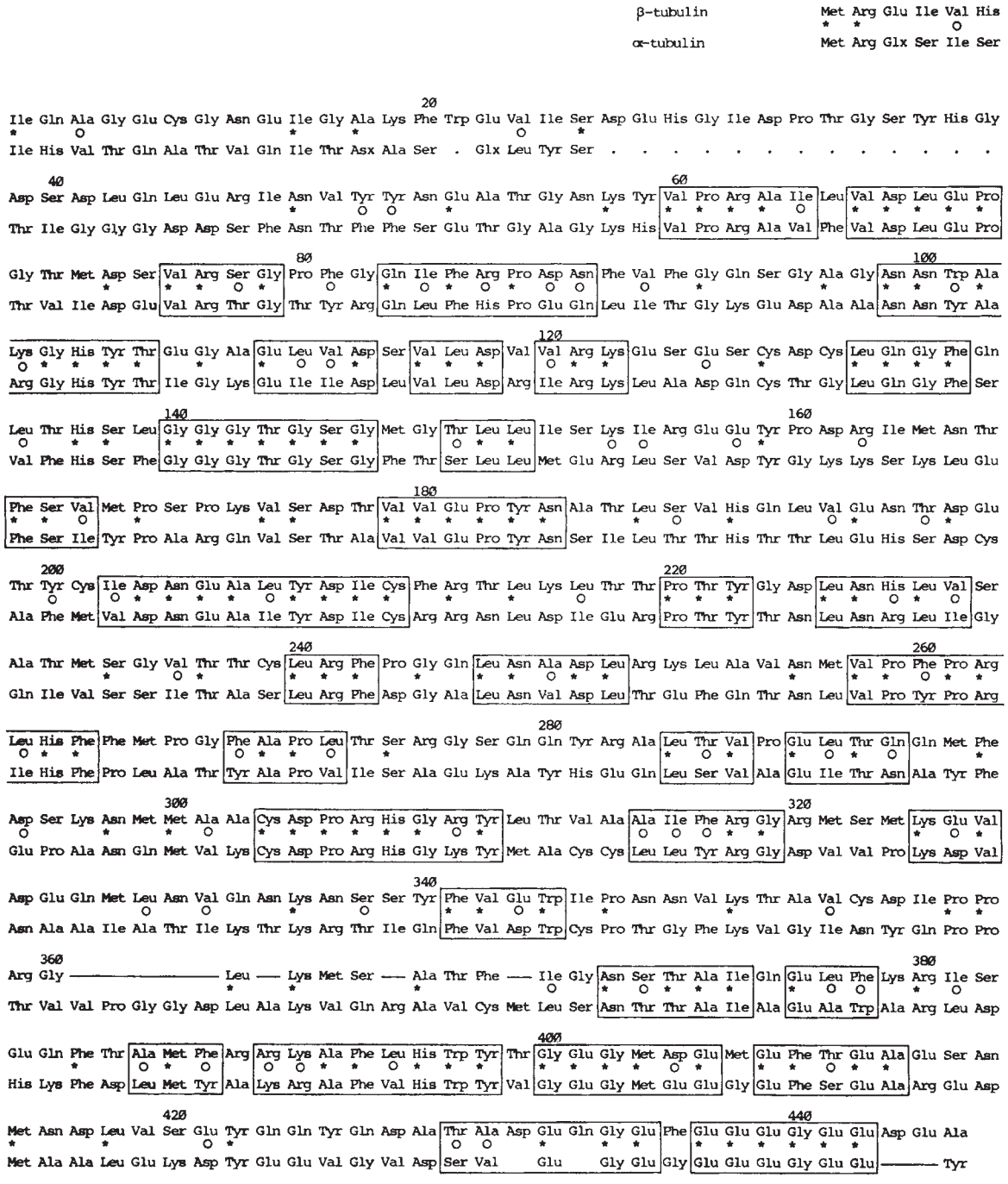

Fig. 5 Comparison of the amino acid sequences predicted from the α and β tubulin mRNAs. The predicted protein sequences for α and β tubulin have been aligned to show regions of homology. Open circles indicate homologous, conservative substitutions. Boxed regions indicate areas of high homology. For completeness, the sequence of the first 25 amino acids of chick brain α tubulin deduced by Luduena and Woodward[5] is also shown. *Identical amino acids in the two sequences.

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event is not the addition but the removal of tyrosine from the C-terminus. This in turn strengthens the argument that there is probably a cyclic process of detyrosylation and tyrosylation and that this may be an important factor in modulating the activity of tubulin in the cell.

Codon usage in α and β tubulin

Figure 4*a*, *b* illustrates the prevalence of codons utilized for each of the amino acids in the translated frames of α and β tubulin, respectively. Surprisingly, strong biases are observed for both tubulins in the case of codons in which the third position is occupied by a G or C. For example, the threonine codons ACC and ACG are used 28 times in both α and β sequences, whereas the ACU and ACA codons are used only twice in either sequence. This contrasts with the reported codon usage for other chicken mRNAs sequenced to date. In ovalbumin mRNA[16], the ACU and ACA codons are used 11 times, but ACC is used only 4 times and ACG not at all. Similarly, for the *src* and *env* proteins of Rous sarcoma virus[17], the known threonine codon usages are 15:18 and 3:6 for ACC+ACG:ACA+ACU. Overall, codons which have G or C in the third position are utilized in the two tubulin sequences 9.6 times more frequently for α tubulin and 8.7 times for β tubulin than codons ending in A or U. The corresponding ratios for ovalbumin, *src* and *env* are 0.9, 2.5 and 1.0. The significance of the disparate codon usage is yet unclear; however, it may reflect some constraint on tubulin mRNA sequences beyond simple conservation of protein sequence.

Homologies between α and β tubulin

The amino acid sequences which we have deduced for α and β tubulin are shown in Fig. 5. The two sequences have been aligned to highlight regions of homology. The tubulins show several interesting features. First, the two sequences are 40–55% homologous throughout, depending on whether exact homology or conservative substitutions are considered. Such homology was first suggested by Luduena and Woodward[5] when they sequenced the N-termini of each chain and found that 11 out of 25 residues were identical. This homology, though distributed among all regions of the sequences, appears in more than 15 clusters of conserved amino acids. For example, the C-terminal regions of both chains contain highly acidic runs of homologous amino acids. Residues 387–411 (numbers are with respect to the β sequence) represent a cluster of 25 amino acids of which all but three are homologous and residues 429–443 comprise a cluster of 14 amino acids of which 11 are homologous. At the nucleic acid level we also find considerable sequence homology (~50%), again with some strongly conserved clusters of nucleic acid residues. An example of this DNA homology can be found in the nucleic acid sequences coding for the amino acids 58–70. Out of 13 amino acids, 11 are homologous in this region; 33 out of 39 nucleic acid residues are identical.

Overall, our sequencing data unambiguously demonstrate that substantial sequence homology exists at both the protein and nucleic acid sequence levels for α and β tubulin. These data confirm the widely held contention that the two tubulin subunits arose from a common ancestral sequence.

As a complementary approach to determining homology we have predicted secondary structure from the amino acid sequences by the methods of Garnier *et al.*[18] and Chou and Fasman[19] for α and β tubulin. With the exception of the C-terminal regions which are both predicted to be highly α helical, additional levels of structural homology were not obvious.

Further comments

The sequence data presented here demonstrate that α and β tubulin share substantial sequence homology and thus probably derived from a common ancestor. There has been speculation that sequence homology might exist between tubulin and muscle proteins. Krauhs *et al.*[20] have reported the location of such homology between actin and α tubulin at residues 22–31 of actin and some residues of α tubulin. This portion of the actin sequence, conserved in all actins sequenced so far[21–26], is ^{22}Ala-Gly-Asp-Asp-Ala-Pro-Arg-Ala-Val-Phe; the corresponding tubulin sequence which we have deduced is ^{57}Ala-Gly-Lys-His-Val-Pro-Arg-Ala-Val-Phe. Thus 7 out of 10 residues in the two proteins, including one stretch of five, are identical. Whether this reflects true homology due to a common binding site for nucleic acid cofactors or a common accessory protein, or whether the homology is merely coincidental remains to be determined by further evolutionary studies.

The hexanucleotide sequence, AAUAAA, has been found ~20 nucleotides from the poly(A) in almost all known polyadenylated RNA sequences and is thought to be the signal for such polyadenylation. However, we find the heptanucleotide, ACAUAAA, located 22 nucleotides 5′ to the beginning of the poly(A) in our β tubulin cDNA. In addition, other mRNAs, like those encoding rat pancreatic amylase[27] and anglerfish pancreatic somatostatin[28], have recently been shown to carry the variant, AAUUAAA. These variations probably reflect flexibility in the sequence requirement of this site.

Heterogeneity of both α and β tubulin chains has been reported many times. This heterogeneity has been observed repeatedly by one- or two-dimensional gel analyses and peptide mapping, and recently by genetic analysis[29,30]. Indeed, we have demonstrated that multiple gene sequences exist for α tubulin and for β tubulin in several organisms (that is, at least four α and four β genes in chicken and *Drosophila*—see refs 3, 4). The availability of the entire protein sequence for one α tubulin and one β tubulin from embryonic chick brain should facilitate additional efforts (1) to localize the regions of heterogeneity of different tubulin subunits and (2) to explore the functional significance of each heterogeneity.

We thank Sonia Bock, David Agard and Dr H. Martinez for their assistance with the computer programs used in the DNA and protein sequence analysis. This work was supported by NIH grants GM 25157 to P.V. and GM 26875 to M.K.

Received 20 October; accepted 2 December 1980.

1. Roberts, K. & Hyams, J. S. (eds) *Microtubules* (Academic, New York, 1979).
2. Kirschner, M. W. *Int. Rev. Cytol.* **54**, 1–71 (1979).
3. Cleveland, D. W. *et al. Cell* **20**, 95–105 (1980).
4. Sanchez, F., Natzle, J., Cleveland, D. W., Kirschner, M. W. & McCarthy, B. J. *Cell* **22**, 845–854 (1980).
5. Luduena, R. F. & Woodward, D. O. *Proc. natn. Acad. Sci. U.S.A.* **70**, 3594–3598 (1973).
6. Lu, R. C. & Elzinga, M. *Biochim. biophys. Acta* **537**, 320–328 (1978).
7. Ponstingl, H., Little, M., Krauhs, E. & Kempf, T. *Nature* **282**, 423–424 (1979).
8. Maxam, A. & Gilbert, W. *Proc. natn. Acad. Sci. U.S.A.* **74**, 560–564 (1977).
9. Feit, H., Slusarek, L. & Shelanski, M. L. *Proc. natn. Acad. Sci. U.S.A.* **68**, 2028–2032 (1971).
10. Cleveland, D. W., Kirschner, M. W. & Cowan, N. J. *Cell* **15**, 1021–1031 (1978).
11. Arce, C. A., Rodriguez, J. A., Barra, H. S. & Caputto, R. *Eur. J. Biochem.* **59**, 145–149 (1975).
12. Argarana, C. E., Arce, C., Barra, H. S. & Caputto, R. *Archs Biochem. Biophys.* **180**, 264–268 (1977).
13. Raybin, D. & Flavin, M. *Biochem. biophys. Res. Commun.* **65**, 1088–1095 (1975).
14. Raybin, D. & Flavin, M. *Biochemistry* **16**, 2189–2194 (1977).
15. Rodriguez, J. A. & Borisy, G. G. *J. Cell Biol.* **75**, 296a (1978).
16. McReynolds, L. *et al. Nature* **273**, 723–728 (1978).
17. Czernilofsky, A. P., Levinson, A. D., Varmus, H. E. & Bishop, J. M. *Nature* **287**, 198–203 (1980).
18. Garnier, J., Osguthorpe, D. J. & Robson, B. *Molec. Biol.* **120**, 97–120 (1978).
19. Chou, P. Y. & Fasman, G. D. *Biochemistry* **13**, 222–245 (1974).
20. Krauhs, E., Little, M., Kempf, T. & Ponstingl, H. *Eur. J. Biochem.* **22**, 285 (1980).
21. Collins, J. H. & Elzinga, M. *J. biol. Chem.* **250**, 5906–5920 (1975).
22. Lu, R. C. & Elzinga, M. *Biochemistry* **16**, 5801–5806 (1977).
23. Vandekerchove, J. & Weber, K. *Proc. natn. Acad. Sci. U.S.A.* **75**, 1106–1110 (1978).
24. Vandekerchove, J. & Weber, K. *Nature* **276**, 720–721 (1978).
25. Gallwitz, D. & Sures, I. *Proc. natn. Acad. Sci. U.S.A.* **77**, 2546–2550 (1980).
26. Ng, R. & Abelson, J. *Proc. natn. Acad. Sci. U.S.A.* **77**, 3912–3916 (1980).
27. MacDonald, R. J. *et al. Nature* **287**, 117–122 (1980).
28. Hobart, P., Crawford, R., Shen, L.-P., Pictet, R. & Rutter, W. J. *Nature* **288**, 137–141 (1980).
29. Kemphues, K. J., Raff, R. A., Kaufman, T. C. & Raff, E. C. *Proc. natn. Acad. Sci. U.S.A.* **76**, 3991–3995 (1979).
30. Kemphues, K. J., Raff, E. C., Raff, R. A. & Kaufman, T. C. *Cell* **21**, 445–454 (1980).

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LETTERS

Can negative molecular ions be detected in dense interstellar clouds?

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The recent laboratory measurement[1-3] of rapid radiative electron attachment processes

$$A + e \rightarrow A^- h\nu$$

where A is a molecular species has renewed speculation on whether negative molecular ions can be synthesized efficiently in dense interstellar clouds. We argue here that for certain interstellar species A, the abundance ratio [A⁻]/[A] may be as high as 0.01–0.1 in commonly assumed physical conditions. If this abundance ratio were correct, negative molecular ions might be detectable in dense interstellar clouds if their microwave spectral frequencies had been determined in the laboratory. It will be shown, however, that this is currently an unlikely prospect.

In recent studies[4-6] of the gas phase chemistry of 'dense' interstellar clouds, the major processes considered in the ambient conditions of low density (10^4–10^6 cm^{-3}) and low temperature (10–50 K) are reactions between positive ions and molecules. These produce polyatomic ions and, on reaction with electrons, a variety of neutral molecules. Cosmic-ray penetration of the clouds produces both an initial assortment of positive ions and electrons. Although the subsequent formation of negative ions through radiative attachment of electrons has been considered[7], it has been concluded that negative ions are both scarce and only a minor contributor to ion–molecule synthetic processes. The scarcity results from the generally slow rate of radiative attachment.

Consider a molecular species A: in dense interstellar clouds, the negative ion A^- can be formed and depleted by the reaction sequence

$$A + e \xrightarrow{k_{ratt}} A^- + h\nu \quad (1)$$

$$A^- + B \xrightarrow{k_2} AB + e,\ C^- + D \quad (2)$$

$$A^- + h\nu \xrightarrow{k_3} A + e \quad (3)$$

$$A^- + I^+ \xrightarrow{k_4} \text{Products} \quad (4)$$

where B and I^+ represent generic neutral and positively-charged species, respectively. If A^- is at steady-state, the abundance ratio $[A^-]/[A]$ is

$$\frac{[A^-]}{[A]} = \frac{k_{ratt} f_e}{k_2 f_B + k_3/n + k_4 f_{I^+}} \quad (5)$$

where n is the total gas density and f_i refers to the fractional abundance of species i. Typical values of k_2 are $\sim 10^{-9}$ cm^{-3} s^{-1} (ref. 8) (independent of temperature) and, of k_4, are $\sim 10^{-7}$–10^{-8} cm^3 s^{-1} at room temperature with $T^{-1/2}$ dependence[9]. The rate coefficient k_3 for photodetachment can generally be neglected in dense clouds for negative ions of several electron volts stability. High pressure, saturated three-body attachment rate coefficients have been studied extensively for stable neutral species[10] and found to vary widely—these values are useful upper limits to the radiative attachment coefficients. For most neutral species, high pressure attachment coefficients are considerably below collision rate coefficients ($\sim 10^{-7}$ cm^3 s^{-1}) at room temperature. High pressure attachment and measured autodetachment rates[10] can lead to an estimate for radiative attachment rate coefficients; such estimates show that for most neutral species these coefficients are quite small at room temperature. Therefore, the recent measurement of 'rapid' ($\sim$0.1–0.01 of the collision rate) room temperature radiative attachment coefficients at low pressure by the ICR technique is indeed intriguing[1,2]. There are no laboratory attachment and detachment data for most interstellar neutral species capable of forming negative ions, thus a theoretical treatment is required. A simple theory for k_{ratt} can be derived from phase space considerations[11-13]. Assuming that no vibrational excitation of the neutral species need be considered, the theory reduces to the following expression for k_{ratt}:

$$k_{ratt}\,(\text{cm}^3\,\text{s}^{-1}) \sim \frac{10^3 h^3 (kT)^{-1/2}}{(2\pi m_e)^{3/2}} \times \frac{g_{A^-}}{2g_A} N_v(\text{EA}) \quad (6)$$

where the computed value of k_{ratt} cannot exceed the (unknown) rate coefficient for temporary electron capture. In equation (6) h is Planck's constant; m_e, the electron mass; N_v(EA), the vibrational density of states of A^- at an internal energy equal to the electron affinity; k, the Boltzmann constant; and g_A and g_{A^-}, the electronic degeneracy factors of A and A^-. Equation (6) is capable of reproducing the large experimental value[1,2] of $k_{ratt} = 2 \times 10^{-8}$ cm^3 s^{-1} at 300 K for SF_6 attachment if, as has been assumed[11], the vibrational frequencies of SF_6^- are half those of SF_6. In general, the lack of known negative-ion vibrational frequencies limits the usefulness of equation (6) even if the statistical assumptions embodied in it are correct.

According to equation (6), k_{ratt} will be large when N_v(EA) is large. The vibrational density of states is a severe function of internal energy and molecular size[12,13] and increases greatly with both increasing electron affinity and molecular size. Based on equation (6) and a knowledge of interstellar chemistry, the most abundant interstellar molecular negative ions stem from neutral free radicals with estimated large (>2eV) electron affinities. For a variety of these species, such as C_4H, C_5N, C_7N, C_9N, C_2H_2CN, C_2H_3O, $C_2H_3O_2$ and possibly C_3N, equation (6) can be used to make plausible radiative attachment rate coefficients near the collision limit of 10^{-7} cm^3 s^{-1} at cloud temperatures of 10–50 K. The first and last radicals mentioned have been detected in clouds[14,15] while the others are possible on synthetic grounds[4-6]. The list has been prepared by removing hydrogen atoms from known stable interstellar molecules. Negative ions formed from such radicals are likely to react with atomic hydrogen via associative detachment processes, for example:

$$C_3N^- + H \longrightarrow IHC_3N + e$$

Assuming that the fractional abundance of atomic hydrogen in dense clouds is $f_B \sim 10^{-4}$ (refs 4–6), k_{ratt} is $\leq 10^{-7}$ cm^3 s^{-1}, k_4 is $\sim 10^{-6}$ cm^3 s^{-1}, k_2 is $\sim 10^{-9}$ cm^3 s^1, and $f_{I^+} \simeq f_e$, we obtain from equation (5) that

$$[A^-]/[A] \leq 10^6 f_e/(1 + 10^7 f_e) \quad (7)$$

Estimates for f_e in dense clouds range from 10^{-7}–10^{-8} (refs 4–6, 16). For $f_e \geq 10^{-7}$, equation (7) predicts $[A^-]/[A]$ to be ≤ 0.1. For the more conservative value of $f_e \sim 10^{-8}$, $[A^-]/[A] \leq 0.01$. Note that for equation (7) to apply, $[A^-] < [e]$.

The major uncertainty in our argument is whether or not radiative attachment rate coefficients near the collision-limited value of $\sim 10^{-7}$ cm^3 s^{-1} occur for only a few measured species[1,2] mostly larger than known interstellar molecules, or whether some smaller radicals with large electron affinities also possess such large values of k_{ratt}. The above simple theory supports the latter possibility. Still, negative molecular ions based on such radicals are not expected to be overly abundant. If we take as

0028–0836/81/080656—02$01.00

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typical an abundance ratio[4-6] [A]/[AH] of $\leq 10^{-2}$ where AH is the stable species (for example, $A = C_5N$, $A^- = C_5N^-$, $AH = HC_5N$), then $[A^-]/[AH] \leq 10^{-3} - 10^{-4}$. Experimental work on the attachment of small free radicals is desirable to determine whether 10^{-3}–10^{-4} is a reasonable estimate or a severe upper limit. If the former assessment is correct, certain negative ions may eventually be detectable in dense interstellar clouds. Also if some relevant radiative attachment rates do approach $10^{-7}\,cm^3\,s^{-1}$, these and related processes must be included in detailed cloud models.

This research was supported by NSF grants AST-7904529 and AST-8020321.

Received 9 October; accepted 12 December 1980.

1. Woodin, R. L., Foster, M. S. & Beauchamp, J. L. *J. chem. Phys.* **72**, 4223–4227 (1980).
2. Foster, M. S. & Beauchamp, J. L. *Chem. phys. Lett.* **31**, 482–485 (1975).
3. Christodoulides, A. A. & Christophorou, L. G. *Chem. phys. Lett.* **61**, 553–557 (1979).
4. Herbst, E. & Klemperer, W. *Astrophys. J.* **185**, 505–533 (1973).
5. Mitchell. G. F., Ginsburg, J. L. & Kuntz, P. J. *Astrophys. J. Suppl.* **38**, 39–68 (1978).
6. Prasad, S. S. & Huntress, W. T. Jr *Astrophys. J. Suppl.* **43**, 1–36 (1980).
7. Dalgarno, A. & McCray, R. A. *Astrophys. J.* **181**, 95–100 (1973).
8. Albritton, D. L. *Atomic Data and Nuclear Tables* **22**, 1–101 (1978).
9. Smith, D. & Adams, N. G. *Top. curr. Chem.* **89**, 1–43 (1980).
10. Christophorou, L. G. *Adv. Electron. Electron Phys.* **46**, 55-129 (1978).
11. Klots, C. E. *Chem. phys. Lett.* **38**, 61–63 (1976).
12. Bass, L., Chesnavich, W. J. & Bowers, M. T. *J. Am. chem. Soc.* **101**, 5493–5502 (1979).
13. Herbst, E. *Astrophys. J.* **237**, 462–470 (1980).
14. Guélin, M., Green, S. & Thaddeus, P. *Astrophys. J. Lett.* **224**, L27–L30 (1978).
15. Guélin, M. & Thaddeus, P. *Astrophys. J. Lett.* **212**, L81–86 (1977).
16. Langer, W. D., Wilson, R. W., Henry, P. S. & Guélin, M. *Astrophys. J. Lett.* **225**, L139–L142 (1978).

Massive neutrinos, helium production and the primordial magnetic field

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Experimental evidence[1] for a non-zero neutrino rest mass, $m_\nu \neq 0$, while still controversial[2], poses intriguing cosmological questions[3-10]. One reassurance is that the possibility of non-zero masses does not necessarily alter previous predictions of the standard big-bang model for the primordial ^{4}He abundance, Y. We show here that a non-zero neutrino rest mass, $m_\nu \sim 10$ eV, may provide the most stringent upper limit proposed to date on the strength of any ordered, primordial magnetic field: $B_0 \leq 4 \times 10^{-9}/m_\nu$ (eV) G. This limit is set by requiring that previous predictions of the standard big bang model for the primordial ^{4}He abundance remain unaltered by the spin procession and subsequent thermalization of right-handed, as well as left-handed, Dirac neutrinos in the early Universe.

Massive neutrinos can be either Majorana or Dirac fermions. Majorana neutrinos are represented by two-component spinor wave functions that are CP eigenstates in the neutrino rest frame. As Majorana mass terms do not conserve lepton number there is no Majorana antineutrino. The total number of spin degrees of freedom per Majorana neutrino flavour is, therefore, $g = 2$, thus the same as for massless neutrinos. Dirac neutrinos, like electrons, are represented by four-component spinor wave functions that are C and P eigenstates in the neutrino rest frame. Dirac mass terms conserve lepton number, so there are both Dirac neutrinos and antineutrinos, and $g = 4$.

For Dirac neutrinos with $m_\nu \neq 0$, both left- and right-handed neutrinos could be present in the early Universe, thereby doubling the number of possible neutrino types. However, neutrino–fermion weak reactions mediated either by vector ($W^\pm$, Z^0) or scalar (Higgs) bosons through the standard Weinberg–Salam–Glashow[11-13] (WSG) model, extended to include Dirac neutrinos with $m_\nu \neq 0$, cannot maintain ν_R in thermal equilibrium at the critical decoupling epoch, $kT_D \sim 1$ MeV (refs 9, 10). In fact, ν_R decouple much earlier at $kT \gg 80$ GeV. Previous studies have shown[14] that, for a present-day baryon density $\rho_B \geq 10^{-2}\rho_c$, any new relativistic particles in the early Universe must decouple at $kT \geq 0.2$ GeV to satisfy the observational constraint on the ^{4}He abundance[15], $Y \leq 0.25$. So, as far as neutrino–fermion weak interactions are concerned, the WSG model extended to include massive neutrinos is consistent with this requirement.

A further consequence of non-zero neutrino rest masses is that neutrinos acquire a magnetic moment ($G_F \equiv 10^{-5} m_p^{-2}$),

$$\mu_\nu = \frac{3eG_F\hbar m_\nu}{8\sqrt{2}\pi^2 c} \tag{1}$$

from diagrams of electro-weak order[16,17]. Consequently, neutrinos can precess in an external magnetic field. Extremely relativistic neutrinos in the early Universe can flip helicities at a rate[16,17]

$$\Gamma^B_{LR} \approx \frac{2}{\pi}\frac{\mu_\nu B_\perp}{\hbar} \approx \frac{3eG_F m_\nu B_\perp}{4\sqrt{2}\pi^3 c} \tag{2}$$

where $B_\perp$ is the component of the primordial field perpendicular to the neutrino velocity. For a sufficiently large magnetic field, $\Gamma^B_{LR} \geq \Gamma_{LL}$, where Γ_{LL} is the rate of neutrino–fermion interactions which thermalize the left-handed neutrinos. For such rapid flipping an equilibrium population of right-handed neutrinos can result, doubling the number of thermal (Dirac) neutrino types (from $N_\nu \geq 3$ to $2N_\nu \geq 6$). This would violate the constraint[14,15] of $N_\nu \leq 4$ imposed on the standard big-bang model by the observed ^{4}He abundance, $Y \leq 0.25$, for $\rho_B \geq 10^{-2}\rho_c$. (If $\rho_B \ll 10^{-2}\rho_c$ the allowed number of neutrino types $N_\nu \gg 4$; a conservative lower limit[5] is $\rho_B \geq 0.006\rho_c$.) Alternatively, satisfying the constraint $N_\nu \leq 4$ by requiring

$$\Gamma^B_{LR} \leq \Gamma_{LL} \tag{3}$$

up to ν_L-decoupling at $T_D \sim 1$ MeV places a surprisingly stringent limit on the strength B_0 of any present-day relic magnetic field.

Our limit is an immediate consequence of three basic assumptions:

(1) The standard big-bang model of nucleosynthesis in the early Universe[18,19] is valid and the primordial ^{4}He abundance satisfies[15] $Y \leq 0.25$.

This assumption requires inequality (3) to hold throughout the nucleosynthesis epoch. Enforcing this requirement at T_D, when $\Gamma_{LL} \sim \Gamma_{exp}$, where

$$\Gamma_{exp} \sim (G\rho)^{1/2} \sim \frac{(kT)^2}{\hbar^{3/2}c^{5/2}}$$

is the expansion rate and ρ is the total density of the Universe, gives

$$m_\nu B_D \leq \frac{4\pi^3\sqrt{2}}{3}\frac{G^{1/2}(kT_D)^2}{eG_F c^{3/2}\hbar^{3/2}}. \tag{4}$$

In equation (4) we have set $B_\perp \sim B_D$, the field strength at neutrino decoupling.

(2) The magnetic field remains 'frozen-in' to the perfectly conducting matter so that $BR^2 \sim$ constant, where R is the expansion parameter. Hence, BT^{-2} does not change as the Universe expands.

Because $\Gamma^B_{LR} \propto B \propto T^2$ while[18,19] $\Gamma_{LL} \propto T^5$, inequality (4) ensures that the population of ν_R cannot equilibrate for any $T > T_D$. Furthermore, if T_0 is the present temperature of the microwave background, then $B_0 = B_D(T_0/T_D)^2$, so inequality (4) gives

$$m_\nu B_0 \leq \frac{4\pi^3\sqrt{2}}{3}\frac{G^{1/2}(kT_0)^2}{eG_F c^{3/2}\hbar^{3/2}} \approx 3.9 \times 10^{-9} \quad \text{eVG} \tag{5}$$

for $T_0 = 2.7$ K (refs 18, 19).

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(3) The neutrino mass probably lies in the range

$$4 \lesssim m_\nu(\mathrm{eV}) \lesssim 20 \tag{6}$$

It has been argued[3-5] that neutrinos with masses in this range would solve the missing mass problem for clusters of galaxies without demanding unacceptably high mass-to-light ratios for individual and binary galaxies and small groups. For this mass range, inequality (5) implies

$$B_0 \lesssim \frac{4\pi^3\sqrt{2}}{3}\frac{G^{1/2}(kT_0)^2}{eG_F m_\nu c^{3/2}\hbar^{3/2}} \approx (0.2\text{–}1.0)\times 10^{-9} \quad \mathrm{G} \tag{7}$$

The right-hand side of inequality (7) is an interesting combination of fundamental microphysical and cosmological constants. Unlike many comparable expressions—such as the large numbers—the sole cosmological parameter, T_0, is not a major source of uncertainty in equation (7).

In deriving inequality (7) we have implicitly assumed that the magnetic field is uniform over all length scales $L \lesssim L_D \equiv ct_D \sim 3\times 10^{10}$ cm at decoupling, implying uniformity over length scales $L_0 \lesssim L_D z_D \sim 30$ pc at present. In the unlikely event that the field is actually tangled over smaller scales $l_0 \ll L_0$ today (and l_0/z_D at decoupling) the upper limit to B_0 is increased over the value given in equation (7) by the random-walk factor $(L_0/l_0)^{1/2} = (30\mathrm{pc}/l_0)^{1/2}$.

Note that inequality (7) provides the most stringent limit on B_0 proposed to date. For example, invoking assumption (2) we find that the requirement that the magnetic energy density does not exceed the radiation energy density only implies

$$B_0 < (8\pi a)^{1/2}T_0^2 \approx 3.2 = 10^{-6} \quad \mathrm{G} \tag{8}$$

for a present black-body radiation temperature $T_0 \approx 2.7$ K. Because $B^2/\rho_B \propto T$, where ρ_B is the baryon mass density, inequality (10) also guarantees that the magnetic energy density remains below the baryon mass density throughout the matter-dominated era. Such a magnetic field is also much too weak to produce an observable anisotropy in the microwave background[20-22]. To guarantee $B^2/8\pi a t^4 < 1$ all the way back to the very earliest epochs, assumption (2), leading to the inequality (8), must be modified by a logarithmic correction factor[23]. Thus, consider a Bianchi type I model with a uniform magnetic field for the anisotropic expansion of a perfectly conducting universe. Requiring that $B^2/8\pi a T^4 < 1$ back to the Planck time $t_p \sim (\hbar G/c^5)^{1/2} \sim 10^{43}$s demands[23]

$$B_0 < (1\text{–}3)\times 10^{-7} \quad \mathrm{G} \tag{9}$$

which is still far less restrictive than equation (7).

For $T \gtrsim 1$ MeV, magnetic field strengths up to $\sim 10^2 \times (8\pi a)^{1/2}T^2 \sim 10^{15}T^2$ (MeV) cause at most a $\sim 15\%$ change in neutron producing and destroying weak reactions involving relativistic electrons and neutrinos[24]. Accordingly, ignoring spin precession, the major effect of a primordial B-field on cosmological nucleosynthesis arises from its influence on the expansion rate and anisotropy of the Universe. Previous limits on B_0 derived by assuming the validity of the standard big-bang model for cosmological nucleosynthesis with $m_\nu \equiv 0$ are therefore equivalent to inequalities (8) and (9).

Finally, if all or part of the Faraday rotation of a distant extragalactic radio source at redshift z can be attributed to a hypothetical ionized intergalactic medium with free electron density $n_{e,0}$ one gets a limit on $n_{e,0}B_0$ if the field is uniform[25-27], and on $n_{e,0}\sqrt{l_0}$ if the field is twisted with a coherence length l_0 (refs 28–30). In the case of a uniform field one obtains a rotation measure[27]

$$\mathrm{RM} = \frac{e^3 n_{e,0}B_{\parallel,0}}{3\pi m_e^2 c^3 H_0 \Omega^2}[(1+\Omega z)^{3/2} - 3(1-\Omega)(1+\Omega z)^{1/2} - 3\Omega + 2] \tag{10}$$

where $\Omega \equiv 8\pi G\rho_0/3H_0^2$, ρ_0 is the total mass density, $H_0 = 100h$ km s^{-1} Mpc^{-1} is Hubble's constant, and $B_{\parallel,0}$ is the line-of-sight component of the field. Observations[30] imply an intergalactic rotation measure $\lesssim 10$ rad m^{-2}, from which one gets (assuming $\Omega z \ll 1$)[25-27]

$$B_0 \lesssim 10^{-10}\Omega_e^{-1}h^{-1} \quad \mathrm{G} \tag{11}$$

for a uniform field out to $z \sim 2$, and

$$B_0 \lesssim 3\times 10^{-9}\Omega_e^{-1}h^{-1/2}[l_0(\mathrm{Mpc})]^{-1/2} \quad \mathrm{G} \tag{12}$$

for a twisted field[30], where $\Omega_e \equiv 8\pi G n_{e,0}m_p/3H_0^2$. Nucleosynthesis calculations, assuming $N_\nu \gtrsim 3$ and $Y \lesssim 0.25$, require $\Omega_B h^2 \lesssim 0.014$, where Ω_B is the present baryon density in units of the closure density[5,15]. Taking $\Omega_e \sim \Omega_B \sim 10^{-2}h^{-2}$, inequalities (11) and (12) imply

$$B_0 \lesssim 10^{-8}h \quad \mathrm{G} \tag{13}$$

for a uniform field, and

$$B_0 \lesssim 3\times 10^{-7}h^{1/2}[l_0(\mathrm{Mpc})]^{-1/2} \quad \mathrm{G} \tag{14}$$

for a twisted field. Smaller electron densities give less stringent limits.

Comparing inequality (7) to inequalities (8), (9), (13) and (14) shows that the most severe constraint on any primordial magnetic field is a simple consequence of a finite neutrino rest mass. Specifically, the calculated magnetic moment[16,17], μ_ν, of massive, four-component Dirac neutrinos in the WSG $SU(2)_L \times U(1)$ theory of weak and electromagnetic interactions[11-13] imposes an upper limit on B_0 an order of magnitude or more lower than any previous limit. (Two-component Majorana neutrinos do not alter the standard nucleosynthesis calculations even when $B_0 \neq 0$ so they set no useful limit to B_0.) Even more stringent limits on B_0 could arise in manifestly left–right symmetric theories with spontaneous parity non-conservation[31], even if the neutrino rest masses are very small, $m_\nu \ll 1$ eV.

We thank G. Feinberg, B. W. Lynn, and K. Olive for informative correspondence, E. E. Salpeter and S. A. Teukolsky for useful discussions, and D. Stewart for aid in preparing the manuscript. This research was supported in part by NSF grant AST 78-20708 at Cornell University.

Received 15 September; accepted 22 December 1980.

1. Reines, F., Sobel, H. W. & Pasierb, E. Preprint (UCI-10 P19–144, 1980).
2. Feynman, R. P. & Vogel, P. *Caltech Note* (1980).
3. Cowsik, R. & McClelland, J. *Phys. Rev. Lett.* **29**, 669 (1972).
4. Tremaine, S. & Gunn, J. E. *Phys. Rev. Lett.* **42**, 407 (1979).
5. Schramm, D. N. & Steigman, G. *Astrophys. J.* (submitted).
6. Bond, J. R., Efstathiou, G. & Silk, J. Preprint (University of California, Berkeley, 1980).
7. Sato, H. & Takahara, F. Preprint (Kyoto University, 1980).
8. Wasserman, I. *Astrophys. J.* (submitted).
9. Dolgov, A. D. & Zel'dovich, Ya. B. Preprint (Space Institute, Moscow, 1980).
10. Shapiro, S. L., Teukolsky, S. A. & Wasserman, I. *Phys. Rev. Lett.* **45**, 669 (1980).
11. Weinberg, S. *Phys. Rev. Lett.* **19**, 1264 (1967).
12. Salam, A. *Proc. 8th Nobel Symp.* (ed. N. Svartholm) (Almquist and Wiksells, Stockholm, 1968).
13. Glashow, S. L. *Nucl. Phys.* **22**, 579 (1961).
14. Steigman, G., Olive, K. A. & Schramm, D. N. *Phys. Rev. Lett.* **43**, 239 (1979).
15. Yang, J., Schramm, D. N., Steigman, G. & Rood, R. T. *Astrophys. J.* **227**, 697 (1979).
16. Lynn, B. W. & Feinberg, G. Preprint (CU-TP-181, 1980).
17. Fujikawa, K. & Shrock, R. Preprint (ITP-SB-80-38, 1980).
18. Peebles, P. J. E. *Physical Cosmology* (Princeton University Press, 1971).
19. Weinberg, S. *Gravitation and Cosmology* (Wiley, New York, 1972).
20. Thorne, K. S. *Astrophys. J.* **148**, 51 (1967).
21. Jacobs, K. C. *Astrophys. J.* **153**, 661 (1968).
22. Jacobs, K. C. *Astrophys. J.* **155**, 379 (1969).
23. Zel'dovich, Ya. B. *Soviet Astr.-AJ* **13**, 608 (1970).
24. Matese, J. J. & O'Connell, R. F. *Astrophys. J.* **160**, 451 (1970).
25. Sofue, Y., Fujimoto, M. & Kawabata, K. *Publ. astr. Soc. Jap.* **20**, 388 (1968).
26. Kawabata, K., Fujimoto, M., Sofue, Y. & Fukui, M. *Publ. astr. Soc. Jap.* **21**, 293 (1969).
27. Brecher, K. & Blumenthal, G. *Astrophys. Lett.* **6**, 169 (1970).
28. Rees, M. J. & Rheinhardt, M. *Astr. Astrophys.* **19**, 189 (1972).
29. Kronberg, P. P. & Simard-Normandin, M. *Nature* **263**, 653 (1976).
30. Kronberg, P. P., Rheinhardt, M. & Simard-Normandin, M. *Astr. Astrophys.* **61**, 771 (1977).
31. Bég, M. A. B., Budny, R. V., Mohapatra, R. & Sirlin, A. *Phys. Rev. Lett.* **38**, 1252 (1977).

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Density evolution of radio sources fuelled by stellar mass loss

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At early cosmological epochs the comoving density of radio sources was very much higher than it is at present. Here we outline a theory for the evolution of the comoving density of radio sources, based on the hypothesis that mass loss from stars flows inwards in massive galaxies to fuel nuclear activity. We use a sufficient condition for the occurrence of inflow to show that the critical galaxy mass above which a total outflow becomes impossible decreases towards earlier epochs. We assume that the comoving density of radio sources is proportional to the comoving density of galaxies with inflow. The predicted density evolution of the radiogalaxy phenomenon is in good agreement with observations.

The evolution of stellar mass loss in galaxies heated by supernovae has been widely discussed[1–5]. In the special case of spherical symmetry, it can be shown[5] that a necessary condition for steady outflow beyond a stagnation radius r_s is

$$\alpha_{sn}E_{sn}(M_T - M_*(r_s)) \geq 2\pi r_s^3 \alpha P_*(r_s) + \alpha M_*(r_s)\left[\frac{GM_*(r_s)}{r_s} + \phi_*(r_s)\right] + \frac{3}{2}\alpha \int_{M_*}^{M_T} \frac{GM_*}{r}\, dM_* \qquad (1)$$

where α_{sn} is the specific mass loss rate due to supernovae, E_{sn} is the energy per unit mass of supernovae ejecta, M_T is the total mass of the galaxy, α is the specific stellar mass loss rate, $P_*(r) = \rho_* V^2/3$ is the effective pressure of the stellar velocity distribution and $\phi_*(r)$ is the gravitational potential. Adopting a de Vaucouleurs $r^{1/4}$ law[6] for the projected mass distribution of elliptical galaxies equation (1) becomes, in dimensionless form,

$$C \equiv \frac{\alpha_{sn}E_{sn}}{\alpha}\frac{r_e}{GM_T} \geq F_{crit}(x) \qquad (2)$$

where $x = r/r_e$ and r_e, is the effective radius. The function F_{crit} is shown for the de Vaucouleurs law in Fig. 1; it decreases monotonically from the value 0.5042 at the centre, to zero at infinity.

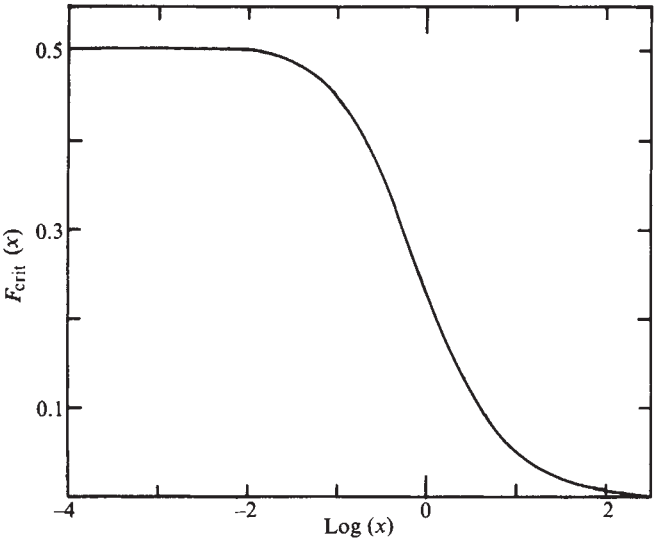

Fig. 1 The wind criterion function $F_{crit}(x)$ for the de Vaucouleurs $r^{1/4}$-law galaxy model is shown versus log (x), where $x = r/r_e$.

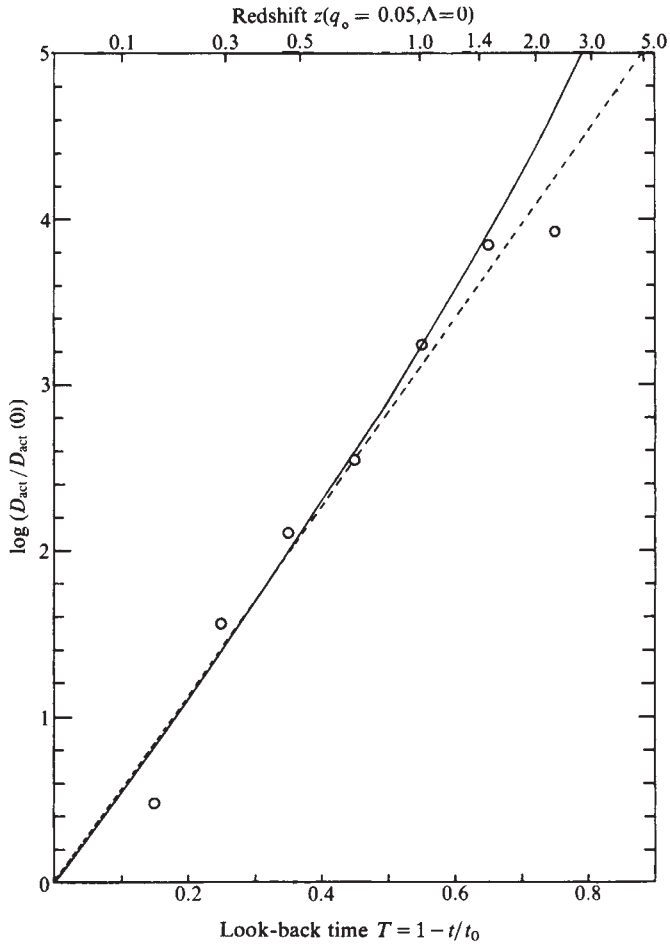

Fig. 2 The predicted comoving density of quasars is shown compared with the observations (○) and approximate analytic form (dashed line) given by Turner[19]. The predicted curve assumes $C_0 = 1.5$, $H_0 = 85$ km s^{-1} Mpc^{-1} and the luminosity function of Kirshner *et al.*[18].

We assume here that inflow occurs whenever conditions are such that total outflow (as indicated by equation (2) with $x = 0$) is not possible. This assumption favours outflowing winds and thus slightly underestimates the proportion of galaxies with inflow.

For practical use it is convenient to rewrite the quantity C on the left-hand side of equation (2) in terms of observable galaxy parameters. The specific mass loss rate $\alpha(t)$ can be obtained from a stellar population model of the galaxy[7]. For a Salpeter initial mass function we find[5] that a good approximation to $\alpha(t)$ is given by $\alpha \simeq 10^{-19}$ $(10^{10}\,\text{yr}/t)\,\text{s}^{-1}$. In principle, the specific supernova mass loss rate α_{sn} could also be obtained from the same stellar population model. In practice, however, uncertainties[8] in the nature of Type I supernovae make such a calculation of dubious validity. Here Type I supernovae are assumed to have low-mass progenitors[9] and are caused by mass transfer in low-mass binary systems[10]. In this case the supernova rate will change on the evolutionary time scale associated with such systems ($\sim 10^{10}$ yr). The difficulties in providing realistic estimates for the evolution of α_{sn} lead us to assume that α_{sn} is independent of epoch, consistent with the expectation of only slow change. Similarly we assume that the mean energy which Type I supernovae inject into the interstellar medium is also independent of epoch. Using the observed supernova rate for elliptical galaxies[11] $\nu_{sn} = 0.157\,h^2$ supernovae per $10^{10}\,L_\odot$ per 100 yr (where $h = H_0/50$ km s^{-1} Mpc^{-1}), and mean supernova energy 4×10^{43} J (ref. 12), we have $\alpha_{sn}E_{sn} = 10^{-8}(L_T/10^{10}L_\odot)\,(10^{11}M_\odot/M_T)h^2\,\text{J kg}^{-1}\,\text{s}^{-1}$.

We now express M_T and r_e/GM_T in terms of galaxy luminosity. An empirical relation connecting the B surface

0028–0836/81/080659—03$01.00

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brightness of a galaxy at r_e with the effective radius r_e has been found by Kormendy[13]:

$$\mu_B(r_e) = \left[3.02 \log\left(\frac{r_e}{1\ \mathrm{kpc}}\right) + 19.74 - 3.02 \log(h)\right] B \text{ mag arc s}^{-2} \quad (3)$$

Combining this with the theoretical relationship between the absolute magnitude of a de Vaucouleurs galaxy, r_e and $\mu(r_e)$:

$$M_B = -39.96 + (\mu_B(r_e)/1 \text{ mag arc s}^{-2}) - 5 \log\left(\frac{r_e}{1\ \mathrm{kpc}}\right) \quad (4)$$

gives $L_B = L_{B,K} h^{x_1} (r_e/1\ \mathrm{kpc})^{x_2}$, where the empirically determined constants are $x_1 = 1.208$, $x_2 = 0.792$ and $L_{B,K} = 9.5 \times 10^{35}$ W.

A second empirical relationship found for elliptical galaxies is the $L - \sigma^4$ correlation, connecting luminosity with observed central line-of-sight velocity dispersion. Using the particular form found by Terlevich *et al.*[14]:

$$\log\left(\frac{\sigma}{1\ \mathrm{km\ s^{-1}}}\right) = -0.0959 M_B + 0.328 + 0.4795 \log(h) \quad (5)$$

we find $L_B = L_{B,T}\, h^{-2}\, (\sigma/100\ \mathrm{km\ s^{-1}})^{x_3}$, where $x_3 = 4.171$ and $L_{B,T} = 7.3 \times 10^{34}$ W. Finally we use the theoretical relation between σ and M_T/r_e, given[15] for small finite apertures by

$$\sigma^2 \simeq 0.2\, GM_T/r_e \quad (6)$$

Expressing B-luminosities in terms of the characteristic luminosity L_B^* which occurs in luminosity functions of the form proposed by Schechter[16], and defining $L_B^* = L_{B,n}^* h^{-2}$, we thus obtain

$$M_T(l) = 1.2 \times 10^{10} \left(\frac{L_{B,n}^{*x_5}}{L_{B,T}^{2/x_3} L_{B,k}^{1/x_2}}\right) h^{-(x_4+2x_5)} l^{x_5} M_\odot \quad (7)$$

where $x_4 = x_1/x_2 - 4/x_3 = 0.566$, $x_5 = 1/x_2 + 2/x_3 = 1.742$ and $l = L_B/L_B^*$. This implies (using our adopted parameter values) $M_T/L_B \propto M_T^{0.43}$ in good agreement with data presented[17] by de Vaucouleurs.

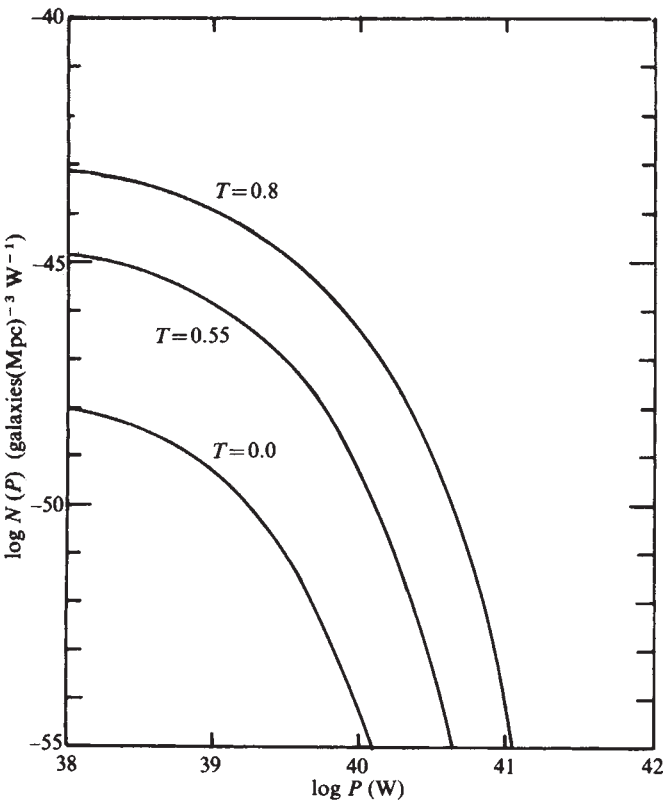

Fig. 3 The predicted co-moving bolometric luminosity function for an accreting black hole model, for three values of the look-back time T.

Combining these relations, the wind criterion equation (2) can be expressed directly in terms of the present value of the dimensionless luminosity l, cosmological epoch t, and other observable quantities. We find, for outflow beyond radius x

$$C \equiv C_0 h^{(2+x_1)/x_2} l^{-(1/x_2+4/x_3-1)} \frac{t}{t_0} \geq F_{crit}(x) \quad (8)$$

where t_0 is the present epoch, and the constant C_0 depends entirely on presently observed quantities such as t_0, $\alpha(t_0)$, $\alpha_{sn}E_{sn}(t_0)$, $L_{B,K}$, $L_{B,T}$, $L_{B,n}^*$ and the ratio L_T/L_B of total-to-blue luminosity. (This arises from the definition[11] of ν_{sn}.) Adopting for this last factor the reasonable value of 10, and taking[18] $L_{B,n}^*$ to correspond to an absolute blue magnitude $M_{B,n}^* = -21.0$, we obtain $C_0 \simeq 1.5$, showing that this coefficient is of the order of unity. The value of C_0, although uncertain, can in principle be determined entirely by independent observations; the time-dependence shown in equation (8) comes from the assumed time-dependence of $\alpha_{sn}E_{sn}/\alpha$ as discussed above.

Inverting equation (8) we find that at epoch $\tau = t/t_0$ all galaxies with luminosity exceeding $l_c(\tau)$ will have non-zero stagnation radii, where

$$l_c(\tau) = (C_0/F_{crit}(0))^{x_6} h^{x_7} \tau^{x_6} \quad (9)$$

and $x_6 = 1/(1/x_2 + 4/x_3 - 1) = 0.819$ and $x_7 = (2 + x_1)x_6/x_2 = 3.316$.

The luminosity function for elliptical galaxies is similar to that of the general field[18]. We therefore assume a Schechter luminosity function defined so that the number of galaxies per unit volume with luminosities in the range $(L, L + \mathrm{d}L)$ is

$$\phi(L)\mathrm{d}L = (\phi^*/L^*)(L/L^*)^a \exp(-L/L^*)\mathrm{d}L$$

The comoving density of galaxies with non-zero stagnation radii normalized to the present epoch $\tau = 1$ is thus given by

$$D(\tau) = n(\tau)/n(1) \quad (10)$$

where

$$n(\tau) = \phi^* \int_{l_c(\tau)}^{\infty} l^a \exp(-l)\, \mathrm{d}l \quad (11)$$

On our model, in which inflowing stellar mass loss fuels active galactic nuclei, we expect the number density of radio sources at any epoch to be proportional to $n(\tau)$. Thus $D(\tau)$ also describes the comoving density of active galaxies, D_{act}.

Observationally it is found[19] that the comoving density of quasars is well fitted by an exponential in look-back time $T = 1 - \tau$, given by $D_{obs} \simeq \exp(13T)$. Figure 2 shows these observations compared with the prediction of our model using parameters $C_0 = 1.5$ (see above), $H_0 = 85\ \mathrm{km\ s^{-1}\ Mpc^{-1}}$ (refs 20, 21) and luminosity function exponent $a = -1.10$ (ref. 18). The predicted curve is insensitive to choice of a, but is more critically determined by C_0, H_0 and $F_{crit}(0)$ through the relation (equation (9)) $l_c(1) \simeq 14$. An equally good fit to the data is obtained for other values of C_0 and H_0 provided $l_c(1)$ is approximately unchanged; for example good fits are also obtained with $(C_0, h) = (0.7, 2), (11.0, 1)$.

Assuming that inflowing stellar mass loss produces nuclear activity with luminosity given by $P = \eta c^2 \alpha(t) M_*(r_s(t))$ (for example, by accretion from a disk onto a massive black hole), we can use the present theory to predict the comoving luminosity function of active nuclei at various epochs. Figure 3 shows the results of such a calculation, made using $\phi^* = 2.35 \times 10^{-3}\ \mathrm{h^3}$ galaxies $(\mathrm{Mpc})^{-3}$ and energy generation efficiency factor $\eta = 0.3$. (The shapes of the comoving function do not depend on either ϕ^* or η.) The predicted luminosity function is in encouraging agreement with that indicated by observations[19], although a detailed comparison is not possible due to the lack of spectral information in such a simple model.

Finally, we have also calculated the numbers of galaxies with inflow as a function of redshift z for an open $q_0 = 0.05$ model

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universe. The predicted counts can be fitted approximately by a powerlaw $N(<z) \simeq 1.5 \times 10^4 z^{5.85}$. The total number of sources out to redshift $z = 4$ is $\sim 5 \times 10^7$, consistent with counts of faint objects[22].

We conclude that the present theory apparently explains the large increase in comoving density of radio sources towards early epochs. At present only massive elliptical galaxies ($L \geqslant 10L^*$) should be radio sources. At earlier times, because of the higher stellar mass loss rate, less massive ellipticals cannot sustain total outflow. The absence of gas in moderately massive ellipticals may thus be a relatively recent phenomenon. On this model the reason for a rapid increase in comoving density of radio sources (by factors $\geqslant 10^3$ over a Hubble time) is principally the steepness of the luminosity function at $L > L^*$. This means that small evolutionary changes in l_c greatly affect the source counts. Whatever the nature of the central source of activity, the present work supports the hypothesis that nuclear activity in elliptical galaxies is caused by inflowing stellar mass loss.

We thank Professor R. J. Tayler and Dr R. C. Smith for helpful comments. This research was supported by the SRC.

Received 17 July; accepted 12 December 1980.

1. Johnson, H. E. & Axford, W. I. *Astrophys. J.* **165**, 381–390 (1971).
2. Mathews, W. G. & Baker, J. C. *Astrophys. J.* **170**, 241–259 (1971).
3. Bregman, J. N. *Astrophys. J.* **224**, 768–781 (1978).
4. Bailey, M. E. *Mon. Not. R. astr. Soc.* **191**, 195–206 (1980).
5. MacDonald, J. & Bailey, M. E. (in preparation).
6. de Vaucouleurs, G. in *Handbuch der Physik* Vol. 53 (ed. Flugge, S.) 311 (1959).
7. Gisler, G. R. *Astr. Astrophys.* **51**, 137–150 (1976).
8. Oemler, A. & Tinsley, B. M. *Astr. J.* **84**, 985–992 (1979).
9. Shklovsky, I. S. *Soviet Astr.* **22**, 413–419 (1978).
10. Whelan, J. & Iben, I. *Astrophys. J.* **186**, 1007–1014 (1973).
11. Tammann, G. A. *Astrophys. Space Sci. Lib.* **45**, 125–130 (1974).
12. Gorenstein, P. & Tucker, W. H. *A. Rev. Astr. Astrophys.* **14**, 373–416 (1976).
13. Kormendy, J. *Astrophys. J.* **218**, 333–346 (1977).
14. Terlevich, R., Davies, R. L., Faber, S. M. & Burstein, D. Preprint (1980).
15. Bailey, M. E. & MacDonald, J. *Mon. Not. R. astr. Soc.* **194**, 195–200 (1981).
16. Schechter, P. L. *Astrophys. J.* **203**, 297–306 (1976).
17. de Vaucouleurs, G. *IAU Symp.* **58**, 1–53 (1974).
18. Kirshner, R. P., Oemler, A. & Schechter, P. L. *Astr. J.* **84**, 951–959 (1979).
19. Turner, E. L. *Astrophys. J.* **231**, 645–652 (1979).
20. de Vaucouleurs, G. *Nature* **266**, 126–129 (1977).
21. Hanes, D. A. *Mon. Not. R. astr. Soc.* **188**, 901–909 (1979).
22. Bohuski, T. J. & Weedman, D. W. *Astrophys. J.* **231**, 653–658 (1979).

Thermal-mechanical properties of a monomolecular film detected by a vibrating reed

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Thin films of organic materials are finding many applications in modern technology. The microelectronics industry, in particular, uses very thin organic films both as photoresists and as insulating layers. The ultimate in organic thin films is represented by molecular monolayers of the order of 2.0 nm thick as deposited by the Langmuir–Blodgett technique[1]. By adapting a vibrating reed apparatus[2] we have investigated the thermal–mechanical properties of a supported 6-nm thick monolayer film. Although a monolayer melting transition could not be found in the resultant spectra, at least two other, low-temperature peaks (at −150 and −20 °C) were detected which seem to be due to relaxation processes both within the film as well as at interfaces of the monolayer structure. Our main aim was to determine whether the mechanical properties of ultrathin films could be measured by the vibrating reed technique, and to detect any thermal–mechanical relaxations which may occur in such films, in particular whether or not a melting transition could be detected at elevated temperatures. Such thermal–mechanical measurements have long been used in the plastics industry to characterize the mechanical properties of resin materials in bulk. The vibrating reed technique may provide an analogous service for very thin organic coatings.

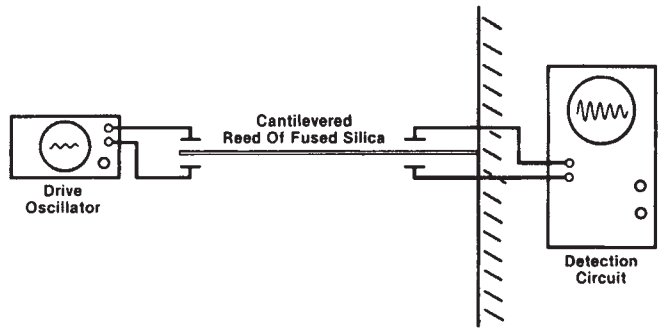

Fig. 1 The vibrating reed apparatus. The silica reed is used as a substrate to support thin films whose mechanical properties are to be investigated.

Other recent work in thin film mechanical spectroscopy has investigated freely suspended liquid crystal films using a disk and support ring apparatus[3–5]. Also the superfluid transition of ^{4}He has been studied using mylar as a substrate[6]. The vibrating reed technique supplements these methods by covering a wide range of temperature and frequency and by not being restricted to monolayer forming films.

Figure 1 gives a schematic representation of the vibrating reed apparatus which is described in detail in ref. 2. The basic concept is: a cantilevered reed is excited into vibration by a set of drive electrodes connected to a suitable oscillator; a corresponding set of detection electrodes, positioned approximately one-third of the way up from the base (a position which avoids setting the electrodes at a vibration node) is used to monitor the amplitude of vibration.

First the reed is excited to a given amplitude and then the drive signal is switched off and the free decay of the reed vibration is monitored. Theoretically, there is an infinite number of modes of vibration into which the reed can be excited by appropriately adjusting the drive frequency. In practice, one works with the first dozen or so lowest tones. The dynamic modulus of the reed material is directly related to the frequency of vibration at any given tone, and the internal friction of the material is inversely proportional to the number of cycles taken for the amplitude to decay to one-half of its initial value. The precise details on measuring these quantities are given in ref. 2.

In the present experiment, a composite sample is used. It consists of a reed-like substrate of fused silica, ~50 μm thick. When operating under a vacuum $\sim 10^{-6}$ torr, the internal friction of the fused silica substrate is exceedingly low over a wide temperature range. This characteristic allows the internal friction of coatings to be detected which may be even 1,000 times thinner than the substrate itself.

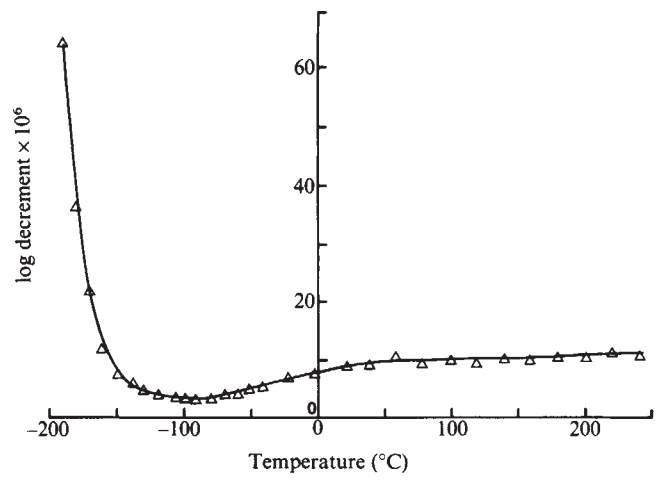

Fig. 2 Plot of internal friction of fused silica from −190 to 240 °C. Note the low and slowly varying behaviour of the internal friction above −150 °C. The solid line was obtained through a piecewise least-square polynomial fit to the data.

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Figure 2 shows the internal friction of fused silica from liquid nitrogen temperature to 240 °C. Note the relatively flat and low internal friction behaviour above −150 °C. Below −150 °C, fused silica shows a large internal friction peak[7] which would make it an unsuitable substrate material in this temperature range. Above −150 °C, however, the internal friction is around 10^{-5}, which implies that typically 10^5 cycles are required for the blank reed to decay to half of its initial amplitude. The solid line in Fig. 2 was used to interpolate the experimental data so that the contribution of the bare substrate to the internal friction of the composite sample could be subtracted accurately and consistently.

An automated Langmuir trough was modified for Langmuir–Blodgett deposition[8]. Constant pressure conditions during dipping were maintained by an electric feedback circuit between the surface pressure monitor and the drive for the movable barrier of the trough. Arachidic acid was spread on an aqueous substrate of 3×10^{-4} M cadmium chloride buffered with 5×10^{-4} M sodium acetate and adjusted to *p*H 6 by the addition of small amounts of acetic acid and/or sodium hydroxide. The film was then compressed to 30 mN m^{-2} and transferred by the Langmuir–Blodgett technique onto a fused silica reed which had been scrupulously cleaned. A film composed of three monolayers of cadmium arachidate[9] on each side of the reed was built up. A fatty acid salt deposited in this way has been shown[10] to be composed of a molecular array in which the alkane chains are close-packed and the metal ions are sandwiched between the carboxyl groups of adjacent monolayers.

Figure 3 summarizes the primary experimental results of this work showing the internal friction of the built-up monolayer film as a function of temperature. The data represent three separate runs where the sample was heated to progressively higher temperatures to see if a melting transition could be detected. Such a transition in cadmium arachidate would be expected to occur at ~140 °C. This expectation is based on an extrapolation from the electron diffraction and frictional data reported by Bowden and Tabor[11]. The high-temperature data, however, showed only a flat baseline behaviour similar to that for the bare reed (see Fig. 2).

We conclude that material from the upper layers of the three-layer film simply sublimed away at the higher temperatures. Therefore no phase transition to a liquid form was observed. However, Fig. 3 clearly shows that there are at least two distinct dispersion peaks at the lower temperatures −150 and −20 °C, respectively. The dramatic decrease in the amplitude of the peak near −150 °C after successive heatings to higher and higher temperatures would seem to indicate that this peak is due to relaxation processes occurring in the bulk of the composite film. The higher temperature peak near −20 °C, however, shows a significantly less decrease in amplitude due to heat treatment. This peak may in fact be due to relaxations occurring at the interfaces of the monolayer structure (either monolayer–monolayer or monolayer–silica). This would also be consistent with the much lower intensity of this peak compared with the one near −150 °C. Figure 4 gives an expanded representation of the mechanical spectrum near −20 °C.

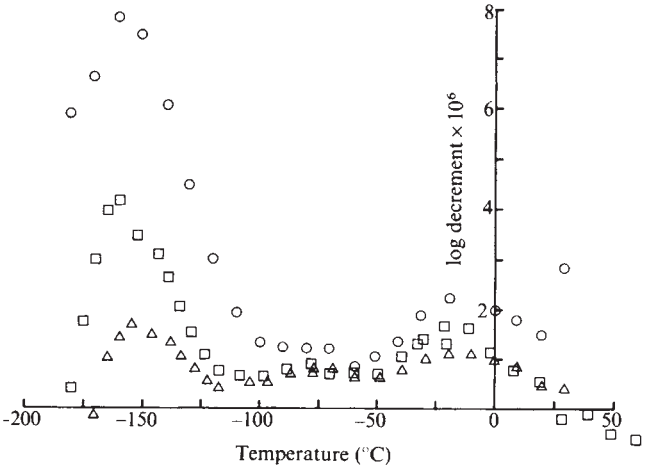

Fig. 3 Internal friction of three molecular layers of cadmium arachidate: ○, heated to 130 °C; □, heated to 160 °C; △, heated to 250 °C. Two dispersion peaks are clearly visible near −150 °C and −20 °C. This plot was obtained by subtracting the internal friction of the bare reed substrate from that of the coated reed. However, a correction factor should be included which depends on the modulus of the substrate and the film and also accounts for the difference in thickness of the two. As there was no way to obtain a reasonable estimate of the modulus of the monolayer film, the data are only proportional to the true internal friction of the film. (See ref. 2 for further details.)

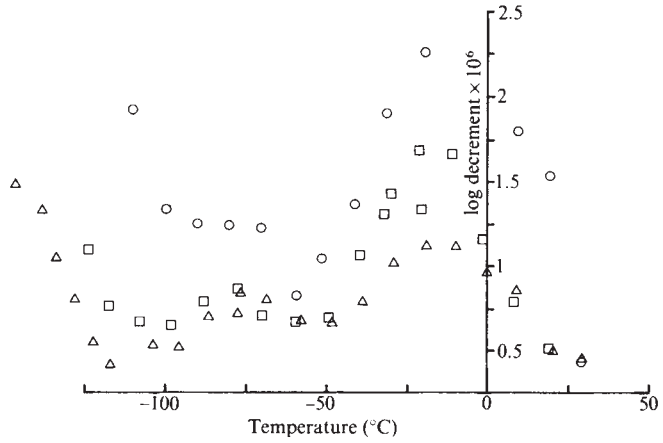

Fig. 4 Expanded view of Fig. 3 near −20 °C. A peak seems to be emerging near −75 °C for the samples which were heated to 160 and 250 °C.

The data in Figs 3 and 4 must be treated with caution as an exceedingly small amount of material is being dealt with, and there are no corroborative data on similar types of samples. However, these data may be clarified by comparison with another physical system which involves long hydrocarbon chains, for example, bulk polyethylene. Low density polyethylene also exhibits two low-temperature transitions near −100 °C and −10 °C respectively. The peak near −100 °C (referred to as the γ peak) seems to occur in polymers with five or more CH_2 units in series and is usually associated with amorphous or interfacial regions in semicrystalline systems. The peak near −10 °C (known as the β peak) is also associated with crystalline imperfections or possibly grain boundaries[12]. These observations support our conclusions if one remembers that our sample film thickness of 6 nm is about the size of an 'interfacial' region in a bulk polymer. Although these conclusions are preliminary, the fact that clear, reproducible spectra can be obtained indicates that the vibrating reed technique has a strong potential as an analytical tool for the study of the mechanical properties of ultrathin films.

We thank B. Berry and W. Pritchet for technical assistance and advice.

Received 10 September; accepted 9 December 1980.

1. Gaines, G. L. Jr in *Insoluble Monolayers at Liquid–Gas Interfaces* Ch. 8 (Wiley-Interscience, New York, 1966).
2. Berry, B. S. & Pritchet, W. C. *I.B.M. J. Res. Devl.* **19**, 334 (1975).
3. Pindak, R., Bishop, D. J., Osheroff, D. D. & Sprenger, W. O. *Paper int. Conf. on Ordering in Two Dimensions*, Lake Geneva, Wisconsin (1980).
4. Pindak, R., Bishop, D. J. & Sprenger, W. O. *Phys. Rev. Lett.* **44**, 1461 (1980).
5. Tarczon, J. C. & Miyano, K. *Paper int. Conf. on Ordering in Two Dimensions*, Lake Geneva, Wisconsin (1980).
6. Bishop, D. J. & Reppy, J. D. *Phys. Rev. Lett.* **40**, 1727 (1978).
7. Anderson, O. L. & Bömmel, H. E. *J. Am. ceram. Soc.* **38**, 125 (1955).
8. Kellner, B. M. J. *Tech. Rep. TR22.* 2326 (International Business Machines Corporation, Hopewell Junction, New York, 1980).
9. Anderson, H. R. Jr. & Swalen, J. D. *J. Adhes.* **9**, 197 (1979).
10. Stephens, J. F. *J. Colloid Interf. Sci.* **38**, 577 (1972).
11. Bowden, F. P. & Tabor, D. *The Friction and Lubrication of Solids*, Part I, 209 (Oxford University Press, 1964).
12. Arridge, R. G. C. *Mechanics of Polymers*, 95–96 (Clarendon, Oxford, 1975).

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Grain boundary microchemistry and stress-corrosion failure of mild steel

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A common cause of failure of large metallic components is stress corrosion, often in subtle combination with fatigue. In the case of carbon steel as used in pipelines, the problem of understanding the cause of variations in susceptibility to intergranular stress corrosion between steel batches of apparently similar origin, history and operating conditions has been researched for many years. We report here the results of investigations including Auger electron spectroscopy which show that the source of variable behaviour is probably the differences in the microchemical content of grain boundary paths, differences which may easily exist between one batch or another of apparently similar steel.

Pure iron is resistant to stress corrosion cracking in nitrate environments, and although attempts have been made to trace the effects of a large number of alloying element additions, such as carbon, aluminium, chromium and nickel, there is apparently little systematic behaviour reported[1,2]. Most previous studies have been carried out on steels containing the usual amounts of the normal individual impurities common to commercial steel stock. Our present investigations suggest that the lack of reproducibility in the reported effects could be due to differences in heat treatment and impurity contents.

Previous Auger electron spectroscopy measurements[3] established that different impurity elements may enrich grain boundaries and other interfaces by factors between 1 and 10^5. Thus, on the assumption (later confirmed experimentally) that impurity elements at low bulk levels do not affect mechanical properties such as yield stress, we undertook a direct study of the microchemistry of the grain boundaries to investigate whether a correlation between susceptibility and bulk impurity content exists.

The test specimens were prepared from 11 ingots of high purity 0.1% C mild steel (Table 1). In 10 of these alloys, the free residual sulphur was tied up chemically and rendered harmless by the addition of 0.05% Mn. To these ingots, a known, small quantity of one commonly occurring impurity element was added separately at that particular bulk level calculated to give rise to a grain boundary segregation content of about 20% of a monatomic layer following an isothermal heat treatment of 100 h at 600 °C. This treatment does not affect the microstructure nor the bulk properties but allows the 20% monolayer segregation to be attained from bulk impurity levels comparable with those encountered in commercial stock. The additions were: 0.3% Al, 0.15% As, 0.01% Ca, 0.1% Cu, 0.3% Ni, 0.03% P, 0.08% Sb, 0.1% Sn and 0.2% Zn. The amount of bulk additive sufficient in each case to produce this 20% monolayer segregation was calculated on the basis of a correlation against maximum solid solubility established earlier[1]. From this, the grain boundary enrichment ratio β_N in a dilute binary alloy is given, within the limits,

$$\beta_N = \frac{1 \text{ to } 10}{X_N} \quad (1)$$

where X_N is the solid solubility limit of element N and β_N is the concentration of the solute in the boundary at equilibrium compared with that in the bulk.

While such quantities were required in advance as part of the experimental design, the predicted segregation levels were later confirmed by exposing the grain boundaries in the spectrometer to direct analysis. In most cases this was difficult, because the levels of impurities were so low that they did not induce sufficient low temperature brittleness to allow exposure of the boundaries by *in-situ* fracture. The successful procedure was to

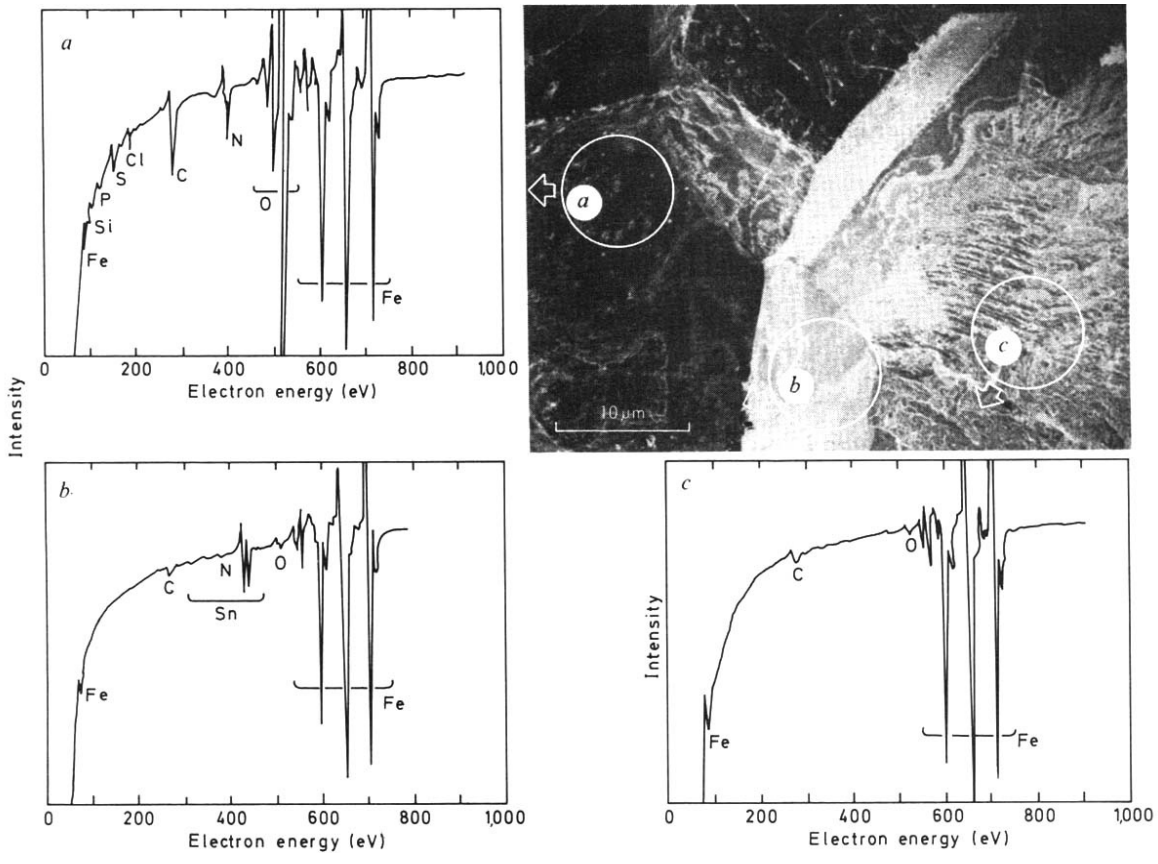

Fig. 1 Auger electron spectra taken from the three regions at a stress corrosion crack front: *a*, the intergranular corroded crack; *b*, the intergranular impact crack; and *c*, the transgranular impact crack.

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Table 1 Analytical results

Cast	Code	Analysed bulk impurity level wt%	Predicted grain boundary content after 100 h at 600 °C (X_B) (monolayers)	Measured grain boundary content (X_B) (monolayers)	Measured corrosive–inert ratio (R)	Relative increase for constant bulk level
1	(S)	0.0026	0.68*	0.80±0.25	0.40±0.05	700±140
2	(pure)	—	—	—	0.86±0.06	—
3	(Al)	0.295	0.18	0.17±0.14	0.57±0.14	1.0±0.3
4	(As)	0.14	0.18	0.19±0.13	0.81±0.17	0.4±0.1
5	(Ca)	0.009	0.15*	0.09±0.07	0.66±0.24	27±11
6	(Cu)	0.10	0.18	0.14±0.05	0.67±0.05	1.9±0.3
7	(Ni)	0.305	0.20	0.10±0.3	0.79±0.12	0.2±0.05
8	(P)	0.028	0.18	0.27±0.13	0.38±0.07	20±5
9	(Sb)	0.076	0.17	0.20±0.07	0.80±0.11	0.9±0.2
10	(Sn)	0.096	0.18	0.26±0.04	0.78±0.09	1.0±0.2
11	(Zn)	0.169	0.15	0.16±0.06	0.81±0.17	0.3±0.1

* Not at equilibrium.

arrest the stress corrosion tensile test before rupture and to transfer the sample from the corrosive solution into the spectrometer, where the fracture was completed by low temperature impact in the ultra-high vacuum. By this process, the pre-existing intergranular stress corrosion crack sometimes extended up to one grain into the virgin steel before the fracture continued in a cleavage mode. These freshly exposed grain boundaries could be isolated and analysed, yielding the actual quantity of segregant on the grain boundaries before stress corrosion failure. Figure 1*a* shows that the stress corrosion crack was heavily oxidized and contaminated by exposure to air and the vacuum environment of CO. Figure 1*b* shows the intergranular fracture surface exposed within the spectrometer and immediately analysed, revealing here a tin segregation and Fig. 1*c*, the remainder of the fracture, which is transgranular and hence shows essentially only iron. The carbon Auger electron signal is associated with monolayer adsorption of CO from the vacuum environment and not with bulk carbon. This is demonstrated by the removal characteristic during ion sputtering of the fracture surface. Using quantification procedures for submonatomic layers, the segregation levels of the added impurities are expressed in terms of fractional adsorbed monolayers, X_B, on the grain boundaries. Table 1 confirms the ~20% level predicted by application of equation (1), within the limits of the equation.

The samples were hot rolled into standard specimen blanks and heat treated in air at 800 °C for 2 h, followed by the isothermal segregation treatment. Flat, unnotched tensile specimens were prepared and tested to failure in an environmental cell of flowing NH_4NO_3 (5M) solution with parallel control tests in a stable environment of liquid paraffin at 80 °C, all at a constant strain rate of $2\times10^{-6}\,s^{-1}$. A similar set of experiments were carried out in constant load testing conditions on a four-point bend rig, submerged in a static bath of the same solution.

The results of the constant strain rate tests are shown in Fig. 2. The error bars in the plotting values indicate the spread in data over six to eight measurements. Every specimen type exhibited stress corrosion in the nitrate environment, failing in a time shorter than that in the control environment. The additions, Zn, As, Sb, Ni and Sn, have a minor effect in reducing rupture lives, Cu, Ca and Al have a modest effect, while S and P clearly have the greatest influence. In the nitrate solution, fracture was predominantly and often entirely, intergranular, while in the control solution, ductile fracture occurred. Table 1 shows the average times to failure in the nitrate solution compared with those in the control solution (corrosive–inert ratio, R). Not only are the yield stresses of all the samples within the range 220 MN m^{-2}±5%, but Fig. 2 shows that the minor impurity additions (as reflected in the times to failure in the inert environment), have not introduced any significant secondary effects on mechanical properties.

We present the results in terms of the change in R, in the analytical form:

$$-\Delta R = 20P + 1.9Cu + 1.0Sn + 0.9Sb + 0.4As + 0.3Zn + 0.2Ni + (700S + 27Ca + 1Al) \quad (2)$$

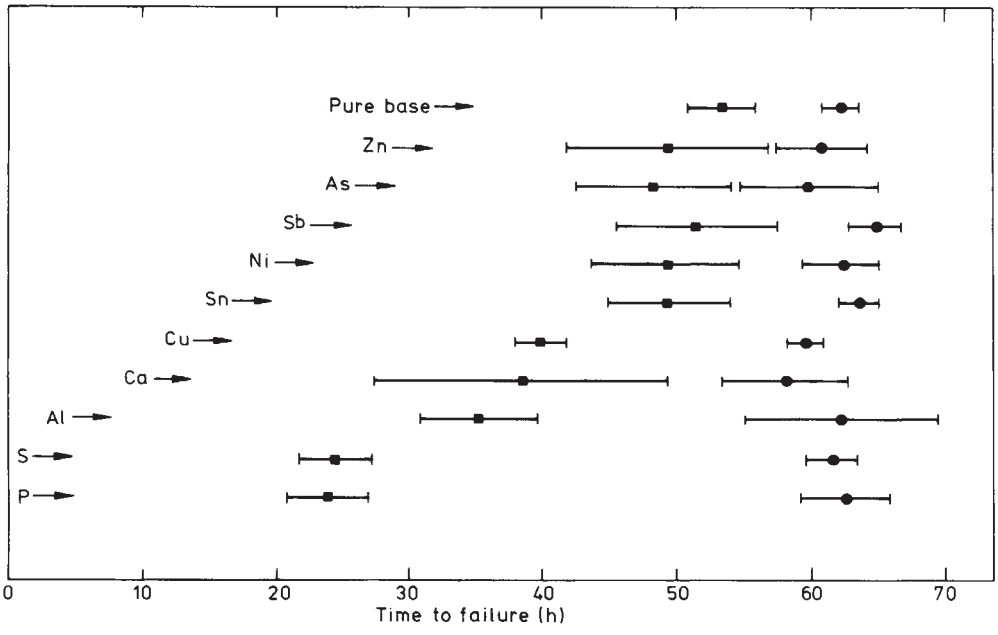

Fig. 2 The fracture times of the 11 alloys stress corroded at a constant strain rate. ●, Tests in paraffin; ■, tests in NH_4NO_3 solution.

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Here, the element symbols represent the bulk content in weight per cent. By introducing the measured enrichment ratio β_N, the changes in R are presented in terms of the bulk concentration of the elements, thus providing a ranking order of potent segregating impurities. In commercial steels, S, Ca and Al are mopped up as precipitates and do not have a segregation role. Thus, to optimize the performance of carbon steels in nitrate environments, ΔR must be minimized. This can be achieved by reducing the bulk impurity levels of the first few elements, but in particular, that of residual phosphorus.

The detrimental influence of phosphorus on the stress corrosion properties of mild steel has been shown by Rädeker and Mishra[4] who added phosphorus in the range 0.012–0.12% to a pure base stock. By using the free energy of segregation obtained from our data, these bulk P levels can be readily transposed to grain boundary segregation levels and the correlation of these X_B values with stress corrosion properties is very similar to that reported here.

We have tried to rationalize the present results in terms of a mechanism whereby a galvanic potential is sustained and the grain boundary is exposed by preferential dissolution of atoms from the boundary. In an oxidizing environment such as aqueous ammonium nitrate, the driving force for atomic dissolution is related to the potential difference between the matrix and the segregants forming a galvanic cell. The hierarchy of the measured differences in the corrosive–inert ratio (ΔR) between each alloy and that of the pure base, normalized to a grain boundary concentration of 1% monolayer, bears a close resemblance to the order of the differences ΔE_0 between the equilibrium potentials of each segregant and that of iron. The potency of each added element for promoting stress corrosion cracking thus increases as $|\Delta E_0|$ increases. The coefficients of other elements, not shown in Fig. 2 or equation (2) can, therefore be predicted approximately as the product of the difference in equilibrium potential from that of iron and the grain boundary enrichment of the element[5]. Thus, although Al, Ti and Ca, for example, all have large values of ΔE_0 they do not segregate, their β values are low, and are therefore not harmful. (Indeed their addition may be beneficial by scavenging those elements which do readily segregate.) Thus, the application of the difference in equilibrium potential plus the grain boundary enrichment factor, seems an adequate first test for sensitivity of a particular element in intergranular stress corrosion.

Received 19 September; accepted 19 December 1980.

1. Parkins, R. N. *J. Iron Steel Inst.* **172**, 149 (1952).
2. Uhlig, H., Perumal, K. E. & Talerman, M. *Corrosion* **30**, 229 (1973).
3. Seah, M. P. & Hondros, E. D. *Proc. R. Soc.* **A335**, 191 (1973).
4. Rädeker, W. & Mishra, B. N. *Werkstoffe Korros.* **21**, 691 (1970).
5. Lea, C., Seah, M. P. & Hondros, E. D. *Mater. Sci. Engng* **42**, 233 (1980).

Controls on low-stress hydro-fracture dilatancy in thrust, wrench and normal fault terrains

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Arrays of parallel extension fractures and veins are sometimes associated with exhumed faults and seem to be the product of repeated hydro-fracturing under a shared stress regime. Here theoretical considerations are used to suggest that this type of hydro-fracture dilatancy is essentially a low differential stress phenomenon. It may develop under hydrostatic fluid pressures around normal faults at shallow depths, but can only occur in association with thrusts when fluid pressures exceed the lithostatic load. This contrasts with most existing models for dilatancy, where the effects only become pronounced at high stress levels and are best developed around thrust faults.

Pre-failure dilatancy, leading to the redistribution of fluids, is a possible cause of observed precursors of shallow earthquakes[1,2], although the extent to which fluid migration is necessarily involved has been questioned[3]. Both the size of the dilatant zone and its stress-dependence are affected by the various types of dilatancy which can occur[4]. However, dilatant effects are commonly attributed to the opening of microcracks under high differential stresses. This accords with the more widespread recognition of earthquake precursors in association with thrust faults, where high stress levels may be characteristically expected[5] and subhorizontal extension cracks may open vertically without restraint[6].

The origin of an observable form of macroscopic fracture dilatancy which is sometimes found around ancient, exhumed faults (Figs 1 and 2*a*) is considered here. The arrays of parallel extension fractures or veins bear a systematic relationship to the faults in accordance with their slip-sense, which suggests that they formed in the same stress field. For example, in Fig. 1, vertical extension veins infilled with fibrous gypsum are associated with parallel-striking normal faults which disrupt a flat-lying sequence of Neogene shales and sandstones at Konarak in south-east Iran[7]. Both sets of structures are compatible with a regime where the maximum principal compressive stress, σ_1, is vertical and the least compressive stress, σ_3, is horizontal and perpendicular to strike. Where such extensional veins abut onto faults they are known as 'feather veins'[8,9]. The vein-fills usually consist of hydrothermal minerals which have grown inwards as fibres from the walls towards a median parting, the texture characteristic of a progressively opening fissure[10]. In many cases, a history of incremental opening is apparent[11], the implication being that fluid flow occurred intermittently with each episode terminating as deposition of hydrothermal minerals destroyed fracture permeability. Self-sealing of fractures in this manner is an important process controlling permeability in active hydrothermal systems[12].

Fig. 1 Conjugate normal faults associated with vertical, gypsum-filled extension veins, Konarak, Makran Coast of south-east Iran. Clastic sandstone dykes (one occurs by the hammer) cut across the gypsum veins before feeding onto the faults.

These textural characteristics, coupled with the typically high length to thickness ratios, suggest that such veins are the product of repeated hydraulic extension fracturing[13]. Similarly oriented barren extension joints may also have formed by hydro-fracturing, but do not retain evidence for more than one episode of opening. Deposition of hydrothermal minerals ensures that some permanent dilatant strain results; at Konarak this locally exceeds 10^{-2} close in to the faults but the veining dies out 100 m or so to each side. From the presence of gypsum, implying temperatures <50 °C (ref. 14), and the observation that at least one of the normal faults displaces overlying Quaternary raised

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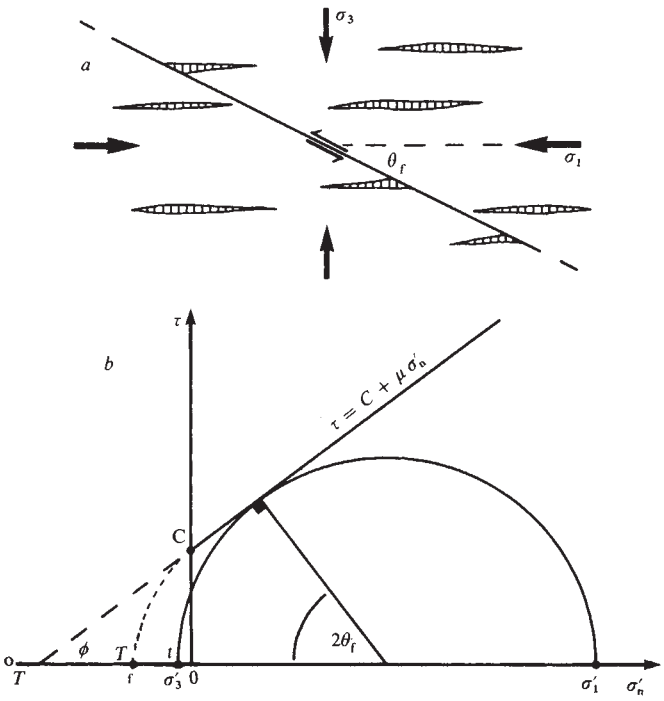

Fig. 2 Hydraulic extension fracturing in association with faults: *a*, field relationships; *b*, critical stress condition for simultaneous shear failure and reopening of extension fractures.

beaches[7], the vein arrays at Konarak have clearly developed in near-surface conditions. However, similar arrays with quartz or calcite as the usual vein filling may occur throughout the frictional regime occupying perhaps the top 10–15 km of quartzo-feldspathic crust[15]. Indeed, Ramsay[11] has attributed extension veins showing multiple increments of opening and sealing (>500 in one case) to repeated hydro-fracturing in upper greenschist facies conditions.

To examine the conditions in which cyclic hydro-fracture dilatancy can occur adjacent to an intermittently slipping fault, the case of an existing fault and extension fracture array in otherwise homogeneous rock is considered (Fig. 2*a*). The simple law of effective stress[16] is assumed to hold so that effective normal stresses are given by

$$\sigma'_n = \sigma_n - P \tag{1}$$

where σ_n is the applied normal stress and P is the pore fluid pressure. Effective principal compressive stresses are thus $\sigma'_1 > \sigma'_2 > \sigma'_3$, and failure criteria can be represented on a Mohr plot of shear stress, τ, versus σ'_n (Fig. 2*b*). In the compressional field, shear failure along the fault is taken to be governed by a frictional criterion of Coulomb form,

$$\tau = C + \mu\sigma'_n \tag{2}$$

where C is the cohesive strength of the fault and the coefficient of friction $\mu = \tan\phi$, ϕ being the angle of friction. The fault is also assumed to be oriented at the optimum angle to σ_1 for frictional sliding, so that $\theta_f = 45° - \phi/2$. Equation (2) may then be rewritten in terms of the effective principal stresses at failure[15] giving

$$\sigma'_1 = 2K^{\frac{1}{2}}C + K\sigma'_3 \tag{3}$$

where $K = ((1+\mu^2)^{1/2} + \mu)^2$. A reasonable value[5] of $\mu = 0.75$ gives $\phi = 37°$, $\theta_f = 27°$ and $K = 4$. The extension fractures or veins are assumed to have a 'healed' tensile strength, t, which is less than the tensile strength of the intact rock, T, and is also small in comparison with C (strictly, $t < \frac{1}{2}C$ for $\mu = 0.75$), so that shear failure is restricted to the compressional field. Thus the condition for reopening the extension fractures is

$$\sigma'_3 = -t \quad \text{or} \quad P = \sigma_3 + t \tag{4}$$

Hydraulic fracturing, requiring $\sigma'_3 < 0$, can only occur when $C > 0$. Thus, hydro-fracturing should not develop adjacent to cohesionless faults. However, natural faults are likely to possess a small cohesive strength through a combination of surface roughness and hydrothermal cementation. Its value is unlikely to exceed 10 bars (ref. 17) unless cementation is marked, in which case it may approach the cohesive strength of intact rock, perhaps 100 bar. If the opening of hydraulic fractures accompanies faulting, an important constraint is placed on the prevailing level of differential stress. From Fig. 2*b* and equations (3) and (4), the critical differential stress for simultaneous shear failure and hydro-fracturing is

$$(\sigma_1 - \sigma_3) = 2(C - \mu t)((1+\mu^2)^{1/2} + \mu) \tag{5}$$
$$= 4C - 3t \text{ for } \mu = 0.75$$

Above this critical stress hydro-fracturing is impossible and only shear failure can occur. Accepting $C = 10$ bar, and taking the extreme case when $t \to 0$, the maximum differential stress allowing hydro-fracturing is just 40 bar. It will be correspondingly greater for intact rocks or a hydrothermally cemented fault, but in comparison with the differential stresses of several kilobars generally required to induce microcrack dilatancy[18], hydro-fracture dilatancy is clearly a low-stress phenomenon.

This critical stress condition may be considered in relation to Anderson's[19] three modes of faulting (based on whether the vertical stress $\sigma_v = \sigma_1$, σ_2, or σ_3), and the ratio of fluid to overburden pressure, $\lambda_v = P/\sigma_v = P/\rho gz$ (ρ being the average rock density, g the gravitational acceleration and z the depth). Previous studies[13,20] have shown that λ_v should be respectively greater and less than unity for hydro-fracturing in thrust and normal fault terrains, but have not taken full account of this limit to differential stress.

For thrust faults $\sigma_v = \rho gz = \sigma_3$, so from equation (4) the condition for hydro-fracturing is

$$\lambda_v = 1 + t/\rho gz \tag{6}$$

requiring $\lambda_v \geqslant 1$ always. In the case of simultaneous hydro-fracturing and normal faulting, $\sigma'_v = \rho gz\ (1 - \lambda_v) = \sigma'_1$, so from equations (3) and (4),

$$\rho gz(1 - \lambda_v) = 2K^{\frac{1}{2}}C - Kt \tag{7}$$

For simultaneous wrench faulting and hydro-fracturing, the particular situation where $\sigma'_v = \sigma'_2 = \frac{1}{2}(\sigma'_1 + \sigma'_3)$ is considered. Then again from equations (3) and (4),

$$\rho gz(1 - \lambda_v) = K^{\frac{1}{2}}C - \tfrac{1}{2}t(K+1) \tag{8}$$

Equations (6), (7) and (8) can be used to separate fields of potential shear failure and hydro-fracturing for the three faulting modes on plots of λ_v versus depth (Fig. 3). As an example, the delimiting curves have been drawn for the top 10 km of a crust with average density $\rho = 2.55$ g cm^{-3}, containing a fault–fracture system with failure parameters, $C = 50$ bar, $t = 10$ bar and $\mu = 0.75$. The barbed side of each curve represents the field of potential hydro-fracturing, except for the thrust regime where the curve marks conditions in which hydro-fracturing must occur. Note that the curve for wrench faulting is displaced towards those for normal and thrust faulting as the value of σ_2 moves respectively towards σ_1 and σ_3 from the median case considered.

Fluid pressures at least equivalent to the hydrostatic head ($\lambda_v = 0.39$ in this case) may generally be expected in established fault zones[15], so that in the uppermost regions of both normal and wrench faults, hydro-fracturing should inevitably accompany faulting. This may account for the extensive fissuring often observed in association with normal faults at the surface[21]. For hydro-fracturing at greater depths, fluid pressures must exceed

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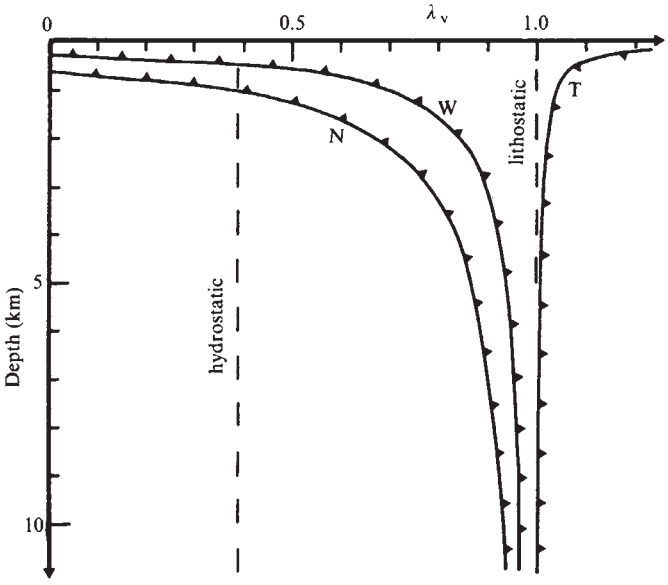

Fig. 3 Curves delimiting potential fields of shear failure and hydro-fracturing (barbed side) for the different faulting modes (T, thrust; W, wrench; N, normal). ($\rho = 2.55$ g cm^{-3}, $\mu = 0.75$, $C =$ 50 bar and $t = 10$ bar).

the hydrostatic head for all faulting modes. Such abnormal fluid pressures may arise through a variety of mechanisms but the presence of highly impermeable 'cap' layers is generally necessary[22]. However, throughout the frictional regime the required λ_v values remain significantly lower for normal faults than for thrusts, with those for wrench faults lying between. I have found that arrays of vertical extension veins around normal faults, as at Konarak, tend to be more common than subhorizontal veins associated with thrust faults. This contrasts directly with the common view[6] that dilatant effects should be more pronounced around thrust faults. Hydro-fracturing (particularly the opening of vertical extension fractures in normal and wrench fault terrains) may clearly also act as a regulatory mechanism, preventing fluid pressure from rising above the critical λ_v value for a given depth and faulting mode, once it exceeds the hydrostatic head.

The relationship between the onset of hydro-fracture dilatancy and the earthquake stress cycle is uncertain, and may vary with faulting mode. However, at Konarak there is evidence for the partial collapse of the dilatant vein array at failure in the form of clastic sandstone dykes which have been mobilized to cut across the gypsum extension veins before feeding onto the faults (Fig. 1). For pre-slip hydro-fracturing during stress accumulation at other than shallow depths around normal and wrench faults, fluid pressure must be sufficiently high for λ_v to exceed or attain the critical value for the particular depth, faulting mode and failure parameters (Fig. 3). Once hydro-fracturing has occurred, fluid pressures should drop towards the hydrostatic, resulting in an increase in effective normal stress and the strengthening of the fault. Shear failure then becomes possible once differential stress has risen sufficiently to overcome this dilatancy hardening effect.

I thank Dr B. K. Atkinson and Dr N. J. Price for helpful comments. Fieldwork at Konarak was accomplished during the 1976 Royal Geographical Society–Imperial College Expedition ot the Iranian Makran. I also thank the expedition members for assistance.

Received 22 September; accepted 8 December 1980.

1. Nur, A. *Bull. seism. Soc. Am.* **62,** 1217–1222 (1972).
2. Scholz, C. H., Sykes, L. R. & Aggarwal, Y. P. *Science* **181,** 803–810 (1973).
3. Mjachkin, V. I., Brace, W. F., Sobolev, G. A. & Dieterich, J. H. *Pure appl. Geophys.* **113,** 169–181 (1975).
4. Nur, A. *Pure appl. Geophys.* **133,** 197–206 (1975).
5. Sibson, R. H. *Nature* **249,** 542–544 (1974).
6. Nur, A., Bell, M. L. & Talwani, P. *Proc. Conf. on Tectonic Problems of the San Andreas Fault System* Vol. 13, 391–404 (Stanford University, 1973).
7. Vita-Finzi, C. *Geol. Min. Surv. Iran Rep.* 47 (1979).
8. Cloos, E. *Proc. natn. Acad. Sci. U.S.A.* **18,** 387–395 (1932).
9. Dennis, J. G. *Am. Ass. petrol. Geol. Mem.* **7,** 95–96 (1967).
10. Durney, D. W. & Ramsay, J. G. in *Gravity and Tectonics* (eds De Jong, K. A. & Scholten, R.) 67–96 (Wiley, New York, 1973).
11. Ramsay, J. G. *Nature* **284,** 135–139 (1980).
12. Grindley, G. W. & Browne, P. R. L. *Proc. 2nd U.N. Symp. on Development and Use of Geothermal Resources* 377–386 (New York, 1976).
13. Secor, D. T. *Am. J. Sci.* **263,** 633–646 (1965).
14. Heard, H. C. & Rubey, W. W. *Bull. geol. Soc. Am.* **77,** 741–760 (1966).
15. Sibson, R. H. *J. geol. Soc. Lond.* **133,** 191–213 (1977).
16. Hubbert, M. K. & Rubey, W. W. *Bull. geol. Soc. Am.* **70,** 115–166 (1959).
17. Hoek, E. & Bray, J. W. *Rock Slope Engineering* (Institute of Mining and Metallurgy, London, 1977).
18. Patterson, M. S. *Experimental Rock Deformation–The Brittle Field* (Springer, Berlin, 1978).
19. Anderson, E. M. *The Dynamics of Faulting* (Oliver & Boyd, Edinburgh, 1951).
20. Hubbert, M. K. & Willis, D. G. *Trans. Am. Inst. Min. Engr.* 210, 153–168 (1957).
21. Abdallah, A. *et al. Nature* **282** 17–23 (1979).
22. Narasimhan, T. N., Houston, W. N. & Nur, A. M. *Geology* **8,** 349–351 (1980).

Reworking in Upper Mesozoic and Cenozoic central Pacific deep-sea sediments

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Data from deep-sea drilling cores from upper Mesozoic and Cenozoic central Pacific Ocean sediments have been used to estimate the frequency with which pelagic microfossils in sediments of different ages include species which have been displaced from older deposits. Intruding microfossils, which include planktonic foraminifera and calcareous nannofossils, and to a minor degree also radiolarians and diatoms, are most easily distinguished by not being contemporaneous with the sediments in which they are found. Presumably their occurrence arises from the mechanical reworking of oceanic sediments by bottom water currents. The data suggest that there were pulses of intensive reworking around 40 Myr BP, 30 Myr BP, 15 Myr BP, 5–10 Myr BP and during the Plio–Pleistocene; these pulses were interrupted by phases of only modest erosion on the Pacific Ocean floor. The absence of reworking of non-contemporaneous fossils in sediments older than 70 Myr suggests relatively sluggish deep-sea currents which were not able to erode deeply into the underlying Cretaceous and in certain areas Jurassic sediments.

Descriptions[1] of sediment columns probed by the Deep-Sea Drilling Project (DSDP) in the central Pacific quite frequently mention the occurrence of non-contemporaneous (reworked) microfossils which are older than the deposits in which they are included. The sites of occurrence (Fig. 1) are distributed over the entire western subtropical and tropical Pacific Ocean, whose basaltic basement is dominantly of Jurassic, Cretaceous and early Tertiary age[2], but there are also sites from which reworked material has not been reported, either because none exists or because it has been overlooked. The collected data[1] come from 35 DSDP sites drilled during 13 legs and are considered representative. Sediments with reworked fossils span a wide range of time (from late Jurassic–early Cretaceous to Quaternary) and depth (>1.5–<6 km), and most of them are located on the Mesozoic part of the Pacific Plate (Fig. 1). The poor coring record during the early DSDP phases and long hiatuses imply that deep-sea basin sites in the central Pacific Ocean are underrepresented in comparison with elevated areas (aseismic rises and ridges). Given the wide area coverage of drill locations there is no bias towards a single deep-sea basin or towards one very special depositional environment. Many of the sites discussed here have been drilled on submarine elevations which in late Mesozoic times were at very shallow water depths, but which subsequently subsided several kilometres. The fact that the displaced fossils have come from pelagic sediments and that most of them have come from a depth interval above the calcite

0028–0836/81/080667—04$01.00
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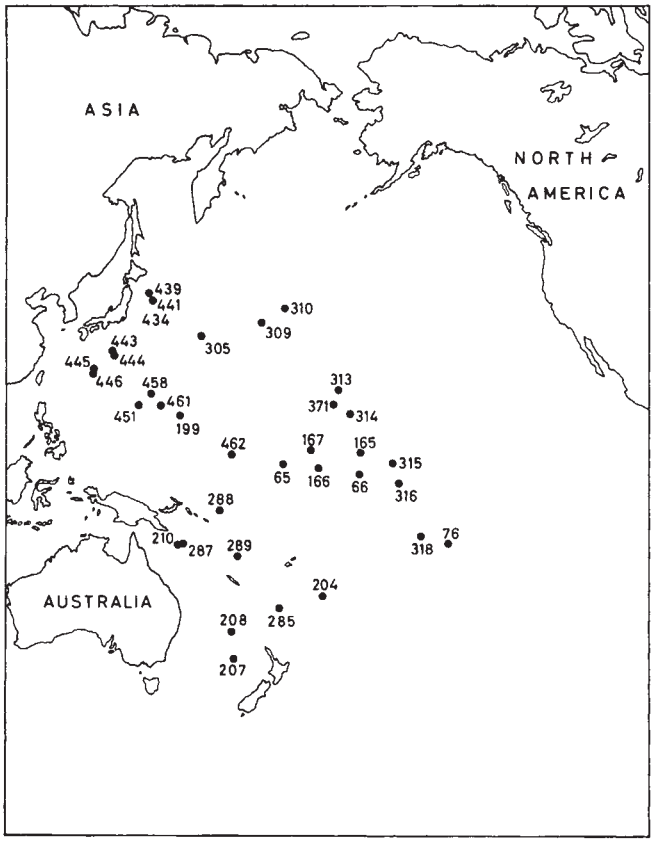
compensation depth (CCD) suggest intermediate (roughly equivalent to bathyal) source regions at the flanks of such submarine highs.

The distribution of displaced pelagic fossils in the central Pacific is based on 207 observations of the four major planktonic microfossil groups (diatoms, radiolarians, coccoliths, foraminifera) whose grain size spectra range from silt to sand-size material. About 80% of the observations are from occurrences of reworked calcareous nannofossils and planktonic foraminifera. The data are therefore biased towards the depositional environments of calcareous oozes above the CCD and under the influence of intermediate[3], but not the deepest, water masses of the oceanic water column.

The biostratigraphic zonations derived from pelagic microfossils are well correlated with absolute chronologies[4–6]. The difference between the age of reworked (zone) fossils and the surrounding sediment can therefore often be determined very precisely. The age difference between the reworked material and the time of displacement (age of the present host sediment) ranges in general between zero (the reworking of contemporaneous or pene-contemporaneous deposits) and a maximum value which is determined by the difference in age of the oldest eroded sediments and the time of deposition of the reworked components. The most surprising feature of the distribution of reworked pelagic fossils in the central Pacific (Fig. 2) is the almost complete lack of redeposition in the Mesozoic. The earliest occurrences are in Maastrichtian sediments, but there are few observations until the late middle Eocene when a major episode of erosion into older deposits began at the depth of the intermediate water masses of the central Pacific. The planktonic forminiferal faunas and calcareous nannofossil floras of many sites bear therefore ample evidence of widespread reworking of Eocene to Cretaceous faunas and floras[7]. The past 40 Myr have been a time of constant reworking of deep-sea sediments (Fig. 2) with peaks of erosive events ~40 Myr BP, 30–32 Myr BP, 14–15 Myr BP, 7–8 Myr BP and during the past 5 Myr. The modest number of observations during 10–13 Myr BP and in particular 15–25 Myr BP and 33–38 Myr BP indicate a temporary decrease of intensity of erosive processes. A characteristic for the central Pacific (Fig. 2) is that only coeval sediments have been reworked during most of the Mesozoic (only Cretaceous).



CENTRAL PACIFIC OCEAN

Fig. 1 Locations of deep-sea drilling sites with good coring records and reworked pelagic fossils in the tropical and subtropical central Pacific Ocean.

The time interval between the age of the eroded section and the time of final deposition of the reworked material can be large and the precision of this variable depends on the stratigraphic resolution[4–6]. The Mesozoic biostratigraphic zonations of deep-sea sediments are considerably less refined than Cenozoic ones[4]; thus Mesozoic and Cenozoic data from the central Pacific cannot be easily compared. However, the complete absence[1] of reworked non-contemporaneous pelagic microfossils in the pre-Maastrichtian parts of the central Pacific sediment section suggest that no older material has been subject to erosion in intermediate water depths during this time span. Age differences deviating considerably from zero only occur since Maastrichtian time (Fig. 2) when fossils from upper Campanian deposits were incorporated into chalks and calcareous oozes. The maximum age difference between the oldest reworked fossil components and the host sediment increased in a regular fashion from close to zero in Maastrichian to ~38 Myr BP in late Eocene time when it reached 30–40 Myr. Between late Eocene and late Pliocene–early Pleistocene the maximum age difference fluctuated between 20 and 70 Myr. Maxima (Fig. 2) developed in the late Eocene, early Oligocene, early to middle Miocene and at the end of the Pliocene. Minima are confined to times when there are few observations of reworked pelagic material. The drastic decrease of the maximum difference in age of the reworked sediment components and the host sediment during early and late Pleistocene observed at Site 462 in the Nauru Basin[8] can also be seen in the data discussed here and seems to be a general feature of the redeposition of older pelagic deposits in the central Pacific.

Reworking and displacement of biogenic deep-sea sediment components imply mechanical erosion and transport of the fossil material[9] from an initial to the final site of deposition. Thus, the occurrence of reworked and displaced fossils in deep-sea sediments provides a simple tool for palaeoceanographic studies. Although reworked fossil material is abundant in certain horizons[10,11], it is generally only a minor fraction of the total sediment. Such fossils are therefore seldom described in detail in studies of the biostratigraphy and/or the depositional environment of Mesozoic and Cenozoic sedimentary sequences. The presence of reworked and displaced contemporaneous pelagic microfossils is often difficult to detect because of the apparently homogeneous nature of many deep-sea sediments and because displaced fossil assemblages cannot easily be discerned from autochthonous ones. The sedimentary structures in the modern ocean floor reflect intense bottom water current activity. The textural properties of deep-sea sediments[12] and sedimentary structures encountered in DSDP cores suggest that this also happened in the past, although it is difficult to evaluate the importance and the amount of reworked coeval material. The presence of displaced, non-contemporaneous pelagic sediment components is much easier to determine than that of contemporaneous material. They offer information about the nature, stratigraphy, palaeodepth of deposition and source of the eroded sediment section as well as about the current regimes of the depositional environments of the host sediment. Calcareous pelagic material has been eroded mostly from intermediate (deeper than neritic, but shallower than CCD) waters, purely siliceous displaced fossil assemblages from regions below the CCD.

The large difference between Mesozoic and Cenozoic redeposition (Fig. 2) of non-coeval reworked pelagic sediment components in the central Pacific is surprising. Maastrichtian events which reworked the directly underlying Campanian sediments represent the beginning of current regimes in the

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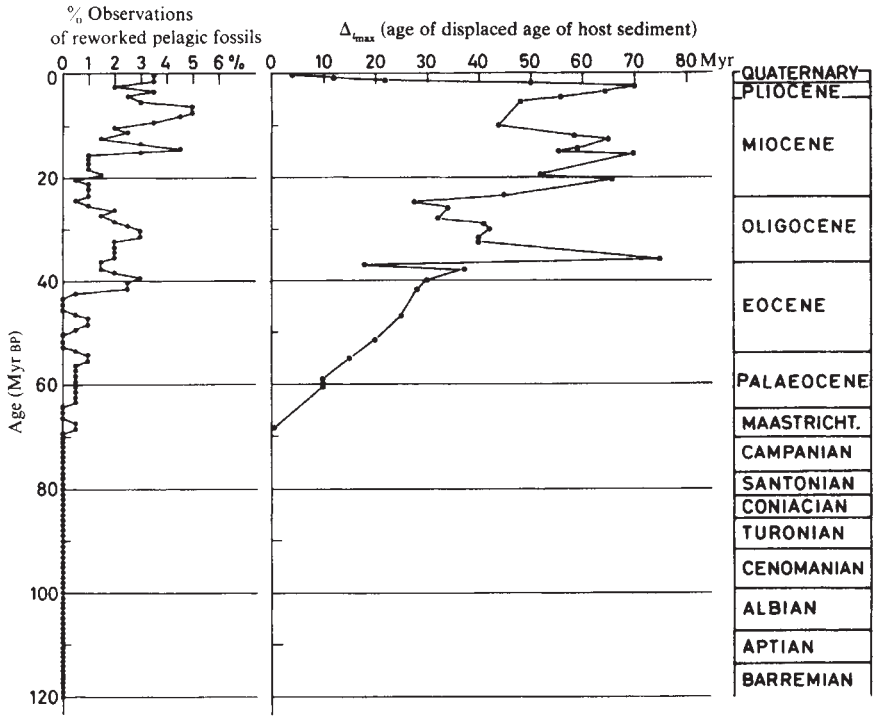

Fig. 2 Occurrence of non-contemporaneous reworked pelagic microfossils (calcareous nannofossils, planktonic foraminifers and radiolarians) in drill sites from the subtropical and tropical central Pacific Ocean (Fig. 1). Occurrence is expressed in per cent of total occurrences of reworked non-contemporaneous microfossils. $\Delta_{f_{max}}$ is the maximum time difference per time interval between the age of the reworked and displaced pelagic fossils and of the host sediment.

intermediate water masses of an intensity which not only allowed transportation and displacement of coeval sediments, but also erosion of the underlying older strata. This is different from the South Atlantic palaeoenvironment where pelagic sediments as old as Coniacian have been displaced into deposits of Santonian age[13]. Although the available data are preliminary, the large quantitative differences of erosion and redeposition during pre-Maastrichtian and Maastrichtian–Pleistocene times indicate a fundamental change of the depositional palaeoenvironment of the deep central Pacific and probably of the world ocean in general. If the physical properties of deep-sea sediments[14] deposited during pre-Campanian time in the central Pacific were not drastically different from those laid down during the past 75 Myr, then their erosive potential should have been approximately the same. Moreover if diagenetic changes have affected the physical properties of the pre-Campanian then these diagenetic changes must have occurred soon after deposition of these sediments.

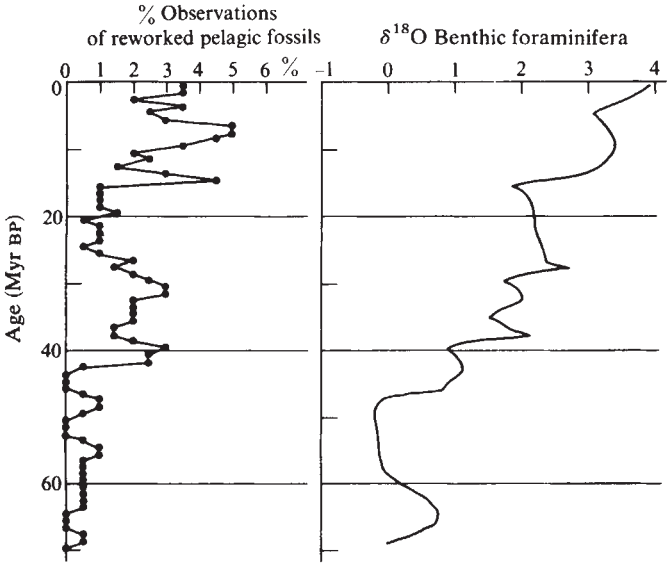

Fig. 3 Occurrence of non-contemporaneous reworked pelagic microfossils in the central Pacific Ocean and oxygen isotope ratios in benthic foraminifers through the past 70 Myr. Isotope data mostly from the North Pacific Ocean[17].

The lack of reworked pelagic microfossils older than their pre-Maastrichtian host sediments coincided with the repeated development of oxygen deficient depositional environments between 80 and 120 Myr BP. The very quiet and sluggish bottom water circulation which is suggested by the laminated sediments[15] of the anoxic lithologies supports the interpretation that a wide area of the pre-Campanian central Pacific in intermediate water depths has not experienced bottom water currents strong enough to erode older strata and to transport the resuspended sediment particles. Relatively rapidly flowing currents seem to have been confined to the upper few hundred metres of the water column, especially close to the equatorial divergence[15].

The number of occurrences of reworked non-coeval pelagic fossils (Fig. 2) remained modest from Maastrichtian to late middle Eocene times, but minor peaks during late Palaeocene and early middle Eocene represent short lived erosive events of a modest scale. The remainder of the Cenozoic was characterized by depositional palaeoenvironments where erosion in intermediate water depths was predominant. The temporal coincidence of important reworking with major changes of the oceanic bottom water temperature regimes (Fig. 3) as deduced from the δ^{18}O ratios of the benthic foraminifers[16,17], suggest that these episodes represent major changes in the bottom water current regimes due to the advection of cold polar probably Antarctic[8,19] to the intermediate waters in the central western Pacific Ocean. This advection resulted in more rapidly flowing currents which eroded the ocean floor and generated widespread hiatuses through reworking and mechanical redeposition of biogenic sediments rather than through dissolution. We do not know whether the pre-late middle Eocene reworking events can be explained by the formation of modest quantities of polar bottom waters much earlier than presumed hitherto which coincided with a significant temperature decrease in high latitudes in Maastrichtian times[20].

The time difference between age of reworked components and time of redeposition reflects the length of the stratigraphic column which was subject to erosion at the time of redeposition; it approached zero during Maastrichtian time, but increased fairly regularly throughout the Cenozoic to the early Pleistocene. The regular increase must mean that the entire sediment column which had been laid down since initiation of the deep

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water current regime in the early Maastrichtian was subject to continuous erosion and redeposition. Deviations from the regular increase of the maximum time difference between the age of the reworked material and of the time of redeposition have been observed during early and late Oligocene as well as during late Miocene and Quaternary. Minima of age difference (Fig. 2) coincide with episodes of high pelagic accumulation rates[21,22], the maxima with the development of important hiatuses in the western Pacific Ocean[23]. The presence of reworked and displaced non-coeval fossils in deep-sea sediments implies mechanical transport and suggests that mechanical erosion rather than non-deposition or dissolution causes most of these hiatuses. The decrease of the maximum age difference between reworked and host sediments in the Central Pacific during the past 3–4 Myr suggests a reduction in the intensity of erosion in the intermediate part of the oceanic water column, despite the deteriorating climate. This is supported by accumulation rate data[24] from the polar South Pacific which indicate a decrease of erosion due to a decrease in current velocities over the past 3 Myr.

Changes of the global palaeoenvironment across the boundary from the late Mesozoic to the Cenozoic era caused: (1) an important decrease in the amount of hydrocarbons which formed on Earth[25]; (2) the initiation of a climatic regime which deteriorated from the equitable late Mesozoic mode to the Glacial mode towards the end of the Cenozoic; and (3) a catastrophy within the biosphere which led to the extinction of many forms of life on land and in the oceanic surface water masses. This development had previously been related to a gradual change of the global palaeoenvironment towards the end of the Cretaceous. More recently instantaneous events such as the sudden injection of brackish water masses from previously isolated Arctic Ocean[26] or the impact of a small celestial body[27] have been invoked to explain the sudden catastrophy at the Cretaceous–Tertiary boundary. However, the evidence from reworked pelagic fossils in the central Pacific Ocean indicates that an important component of the late Mesozoic palaeoenvironment, the current regimes of the intermediate and deep oceanic water masses, changed ~5 Myr earlier than the catastrophic events on land and in the pelagic surface waters of the oceans.

The data used in this report have been produced by the shipboard palaeontologists and sedimentologists of the DSDP Legs 7, 9, 17, 20, 21, 30, 32, 33, 57 to 61, their skilful work is acknowledged. We thank O. Eldholm (Oslo), and W. H. Berger for helpful comments. The Deep-Sea Drilling Project (DSDP) is operated by Scripps Institution of Oceanography with advice from the Joint Oceanographic Institutions.

Received 9 July; accepted 4 December 1980.

1. Thiede, J., Boersma, A., Schmidt, R. R. & Vincent, E. *Init. Rep. DSDP* Leg **62** (in the press).
2. Pitman, W. S., Larson, R. L. & Herron, E. M. *Magnetic Lineations of the Oceans, Map* (Geological Society of America, Boulder, 1974).
3. Reid, J. L. *The Johns Hopkins Oceanogr. Stud.* **2**, (1965).
4. van Hinte, J. E. *Am. Ass. petrol. Geol. Bull.* **60**, 498–516 (1976).
5. Hardenbol, J. & Berggren, W. A. *Am. Ass. petrol. Geol. Stud. Geol.* **6**, 213–234 (1978).
6. Berggren, W. A. & van Couvering, J. A. *Palaeogeogr. Palaeoclimatol. Palaeoecol.* **16**, 1–216 (1974).
7. Krasheninnikov, V. *Init. Rep. DSDP* Leg **62** (in the press).
8. Schlanger, S. O., Premoli-Silva, I., Rea, D. K. & Thiede, J. *Init. Rep. DSDP* Leg **61** (in the press).
9. Seibold, E. *Micropaleontology* **24**, 407–421 (1978).
10. Beckmann, J. P. *Init. Rep. DSDP* Leg **33**, 467–470 (1976).
11. Kaneps, A. G. *Init. Rep. DSDP* Leg **33**, 361–367 (1976).
12. van Andel, T. H. *J. Geol.* **81**, 434–457 (1974).
13. Premoli-Silva, I. & Boersma, A. *Init. Rep. DSDP* Leg **39**, 615–641 (1977).
14. Schlanger, S. O. & Douglas, R. G. *Int. Ass. Sediment. Spec. Publ.* **1**, 117–148 (1974).
15. Dean, W., Claypool, G. & Thiede, J. *Init. Rep. DSDP* Leg **62** (in the press).
16. Savin, S. M. *Ann. Rev. Earth planet. Sci.* **5**, 319–335 (1977).
17. Vincent, E. & Berger, E. H. in *The Sea* Vol. 7 (ed. Emiliani, C.) (Wiley-Interscience, New York, in the press).
18. Shackleton, N. J. & Kennett, J. P. *Init. Rep. DSDP* Leg **29**, 743–755 (1975).
19. Keigwin, L. D. *Nature* **287**, 722–725 (1980).
20. Hecht, A. D. *Foraminifera* **2**, 1–43 (1976).
21. van Andel, T. H., Heath, G. R. & Moore, T. C. *Geol. Soc. Am. Mem.* **143**, 1–84 (1975).
22. Worsley, T. R. & Davies, T. A. *Science* **203**, 455–456 (1979).
23. Moore, T. C., van Andel, T. H., Sancetta, C. & Pisias, N. *Micropaleontology* **24**, 113–138 (1978).
24. Ledbetter, M. T. & Huang, T. C. *Mar. Geol.* **36**, M21–M28 (1980).
25. Irving, E., North, F. K. & Couillard, R. *Can. J. Earth Sci.* **11**, 1–17 (1974).
26. Thierstein, H. R. & Berger, W. H. *Nature* **276**, 461–466 (1980).
27. Hsü, K. J. *Nature* **285**, 201–203 (1980).

Cenozoic palaeotemperature changes and planktonic foraminiferal speciation

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The interaction of plate movements and progressive climatic deterioration produced a constantly changing oceanic environment during the Cenozoic. The destruction of the Tethyan Seaway and the severing of low-latitude oceanographic connections during the past 65 Myr (ref. 1), coupled with the initiation of unrestricted circum-Antarctic circulation in the middle to late Oligocene[2], provided the necessary boundary conditions for the development of a global ocean marked by steep latitudinal temperature gradients. This is in contrast to the rather mild climate of the Mesozoic, which was characterized by an ocean with a homogeneous thermal structure and weak latitudinal temperature gradients[1]. Throughout the Cenozoic, marine plankton were constantly adapting to major changes in the oceanic environment. This can be seen, for example, in the continual development of latitudinal provincialization as a response to polar coolings[1,3]. Here I attempt to evaluate the relationship between palaeotemperature change and planktonic foraminiferal speciation during the past 65 Myr.

Earlier studies compared patterns of planktonic foraminiferal evolution with available palaeotemperature data[4–8]. Frerichs[7] suggested that rapid radiation and high diversity of planktonic foraminifera coincided with periods of warm climate, whereas Cifelli[5] and Lipps[6] both concluded that planktonic foraminiferal diversity was closely regulated by high-latitude marine climates. According to their models, when latitudinal temperature gradients are enhanced there is increased provincialization, while periods with globally uniform oceans are marked by low diversity and wide geographical distribution of species. Stehli *et al.*[9] and Cifelli[5] also proposed that the rate of evolution of new taxa is highest in regions of warmest surface waters. If true, marine sequences from tropical–subtropical regions should be an excellent record of the major features of planktonic foraminiferal evolution. Hart[10] suggested that not only is foraminiferal evolution closely related to palaeotemperature changes, but that the interative trends[5,11] observed during the Cenozoic "indicate successive attempts at colonizing deeper levels of the winter column".

The present data are based on a synthesis of Palaeocene to Recent planktonic foraminiferal distributions in marine and land-based records from the Caribbean region[12]. This compilation is based primarily on the results of Legs 4 (ref. 13) and 15 (refs 14, 15) of the Deep Sea Drilling Project, as well as information from the Antilles and northern South America[16–18]. The past 65 Myr has been divided into 43 planktonic foraminiferal zones[14,15], varying in duration between ~0.5 and 3.4 Myr (ref. 19). For each of these zones species diversity and the number of first and last appearances have been determined (Fig. 1). The species diversity data enable the total rate of change of planktonic foraminifera to be estimated in terms of number of species per Myr (Fig. 1).

The $\delta^{18}O$ record used in this study is a compilation of previously published benthic foraminiferal data[20]. The Cenozoic $\delta^{18}O$ record reveals that there has been a persistent cooling of the oceans since the late Palaeocene. Superimposed on this trend are a series of distinct climatic and oceanographic events, including: (1) the development of the psychrosphere near the Eocene–Oligocene boundary[21]; (2) the establishment of an unrestricted circum-Antarctic current in the middle to late Oligocene[2]; (3) the development of a permanent Antarctic ice cap in the middle Miocene[22]; and (4) the onset of Northern Hemisphere glaciation in the late Pliocene[23–26].

0028–0836/81/080670—03$01.00

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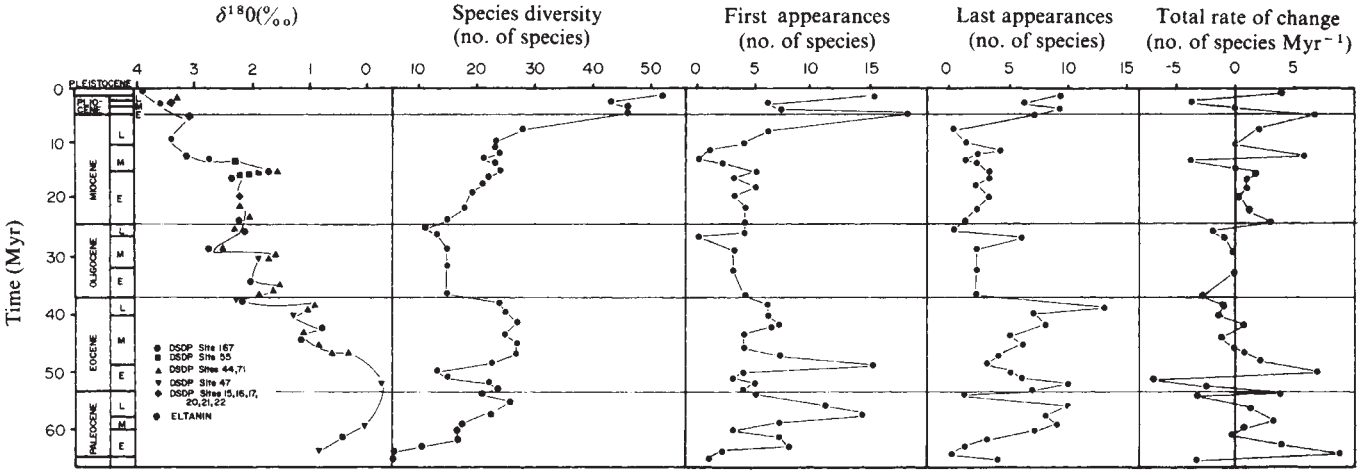

Fig. 1 Comparison of Cenozoic $\delta^{18}O$ record with various faunal parameters reflecting the diversity of planktonic foraminifera. The $\delta^{18}O$ curve is from ref. 20 and is a composite record based on analyses of benthic foraminifera from different deep-sea sedimentary sections. The faunal data base is from ref. 12. Geological epochs and an absolute time scale after Berggren[19] are plotted on the left.

The palaeotemperature record indicates that a gradual warming occurred throughout the Palaeocene. Associated with this warming is an increase in species diversity (from <5 species at the base of the Palaeocene to >25 species in the late Palaeocene), with the morphologically simple globigerine forms that survived the massive extinctions at the end of the Mesozoic giving rise to the more advanced globorataliid forms[5,27]. There are two distinct episodes of rapid origination within the Palaeocene, one in the early and one in the late Palaeocene. These are reflected in the total rate of change of the fauna, with the Palaeocene generally being a time of net gain in overall number of species.

Following the diversity high in the late Palaeocene there was a rapid decrease in species number with a low being reached by the end of the early Eocene. There is a sharp decline in the number of first appearances during this time interval, while the number of last appearances remains relatively high. The result is a net loss in species with the latter part of the early Eocene being marked by the greatest decrease for the entire Cenozoic. This is when the long-term cooling trend for the Cenozoic probably began, although there is no abrupt change in the isotopic record with which we can specifically correlate the observed faunal events.

A second major increase in species diversity occurred in the Palaeogene, extending from the middle Eocene through the end of the Eocene, where first appearances exceed extinctions. The major evolutionary feature during this period was the introduction of hastigerine, orbuline and truncorotaliid forms[5,27]. The palaeotemperature record indicates a brief warming during the middle Eocene followed by relatively stable climatic conditions throughout the remainder of the Eocene. (A similar pattern is shown by isotopic work reported in refs 22, 28, 29.)

One of the most dramatic events in the Tertiary oxygen isotopic record is associated with the Eocene–Oligocene boundary (~38 Myr ago). This rapid isotopic change has been attributed to a cooling of Antarctic waters and the development of cold bottom water circulation (psychrosphere) similar to that of the present day[21]. The response of planktonic foraminifera to this new oceanographic regime is manifested as a sudden drop in species diversity, (from ~25 to 15 species) with the largest number of last appearances for the entire Cenozoic. By the end of the Eocene, all globorotaliid, hastigerine, orbuline and truncorotaliid forms had become extinct. As at the Cretaceous–Tertiary boundary, the simple globigerine forms were among the few survivors of the Eocene–Oligocene event. A similar crisis in benthic foraminifera was reported by Kennett and Shackleton[21], although Corliss[30] suggests that the benthic turnover may have occurred in more gradual steps.

Following the climatic deterioration at the Eocene–Oligocene boundary, fairly stable oceanographic conditions were maintained until the middle Oligocene. This stability is reflected in a planktonic foraminiferal fauna with uniformly low diversity. The simple globigerine assemblage present at this time has a global geographical distribution indicating the absence of any strong latitudinal thermal barriers.

Another abrupt isotopic shift occurs in the middle Oligocene. This isotopic event coincides with the initiation of unrestricted circum-Antarctic flow[2], and may reflect a high-latitude cooling. This isotopic event coincided with the largest eustatic drop in sea level of the entire Cenozoic[31]. Each faunal parameter suggests that there was a delay in the response of planktonic formaminifera to this oceanographic event. Shortly after the isotopic shift, planktonic foraminiferal species diversity reached a low point, the number of first appearances dropped to zero, and there was an increase in last appearances. This offset in the isotopic and faunal records may reflect a lag in the response of planktonic foraminifera to changing surface conditions, or it may be a stratigraphic artefact. The ubiquitous globigerine forms continue to dominate the planktonic foraminiferal fauna of the middle and late Oligocene.

This diversity low in the late Oligocene is followed by a very uniform increase in the number of planktonic foraminiferal species that begins in the early Miocene and continues throughout the remainder of the Neogene. Species diversity increases from ~10 species in the late Oligocene to 25 species by the end of the early Miocene. Cifelli[5] considered this to be one of two major expansions in planktonic foraminifera that occurred during the Cenozoic (the other following the massive extinctions at the Cretaceous–Tertiary boundary), with the modern fauna beginning to evolve at this time. Phylogentically different lineages of globigerinoids, globorotaliids, hastigerines and orbulines reappeared during the early Miocene. The palaeotemperature record indicates a slight warming at the end of the Oligocene, followed by stable conditions during the early Miocene. Associated with these uniformly warm conditions of the early Miocene were a constant rate of first appearances and a low level of last appearances resulting in net gain in total fauna.

The relatively stable oceanographic conditions of the early Miocene ended abruptly in the middle Miocene (about 14–12 Myr ago) with the development of a permanent Antarctic ice cap[22]. This major climatic event is manifested by the largest isotopic shift in the Cenozoic record, and further enhanced latitudinal provincialization of all planktonic microfossil groups[3]. First appearances drop to zero, while the number of extinctions increases slightly in the middle Miocene producing a brief but relatively large net loss in species. Throughout this period species diversity remains fairly stable, fluctuating between 21 and 24 species.

The most rapid increase in species diversity during the past 65 Myr occurred in the early Pliocene. Within several million

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years, the number of planktonic foraminiferal species increased from <30 in the late Miocene to >45 in the early Pliocene. Since the middle Miocene, the world had been in a glacial state but the isotope record indicates that a slight retreat of the Antarctic ice cap may have occurred in the early Pliocene. The rapid increase of planktonic foraminifera at this time may have been a response to this brief amelioration.

The most recent major climatic threshold was the initiation of Northern Hemisphere glaciation in the late Pliocene (~3.0 Myr ago)[23-26]. This probably intensified existing latitudinal temperature gradients. This climatic change immediately caused a dramatic drop in number of first appearances, a slight decline in species diversity and a short-term net loss in species.

Since the inception of the northern ice sheets, there have been periodic oscillations between glacial and interglacial climates. However, these rapidly changing climatic conditions have had very little adverse effect on planktonic foraminifera. The general pattern has been for the geographic ranges of individual species to expand and contract in response to changes in latitudinal temperature gradients, without any appreciable loss of species. Once planktonic foraminifera became adapted to the effects of a Northern Hemisphere ice sheet, species diversity increased and reached a maximum for the entire Cenozoic (>50 species).

This comparison of the Cenozoic palaeotemperature record and species diversity in planktonic foraminifera confirms the previously reported[4,7,8] close relationship between palaeoclimatic change and planktonic foraminiferal evolution. During the past 65 Myr, there have been five periods during which there was a net loss of the total planktonic foraminiferal population. These periods of net loss correspond closely to the major climatic events of the Cenozoic, and include the initial phase of climatic deterioration in the early Eocene, the development of the psycosphere near the Eocene–Oligocene boundary, the initiation of unrestricted circum-Antarctic flow in the middle to late Oligocene, the build-up of a permanent Antarctic ice cap in the middle Miocene and the onset of Northern Hemisphere glaciation in the late Pliocene. Following each of these major climatic events there was generally an expansion of planktonic forminifera associated with milder climate. The species diversity record clearly shows that there were two major radiations during the Cenozoic, one beginning in the early Palaeocene and one beginning in the early Miocene[5].

I thank Drs W. A. Berggren, B. H. Corliss and J. P. Kennett for helpful comments. This work was partially supported by NSF grant OCE79-23736 and a Research and Productive Scholarship Grant from the University of South Carolina.

Received 15 September; accepted 4 December 1980.

1. Berggren, W. A. & Hollister, C. D. *Soc. Econ. Paleont. Mineral., Spec Publ.* **20**, 126–186 (1974); *Tectonophysics* **38**, 11–48 (1977).
2. Kennett, J. P. *J. geophys. Res.* **82**, 3843–3860 (1977).
3. Kennett, J. P. *Mar. Micropaleont.* **3**, 301–345 (1978).
4. Berggren, W. A. *Micropaleontology* **15**, 351–365 (1969).
5. Cifelli, R. S *Syst. Zool.* **18**, 154–168 (1969).
6. Lipps, J. H. *Evolution* **24**, 1–22 (1970).
7. Frerichs, W. E. *J. Paleont.* **45**, 963–968 (1971).
8. Fischer, A. G. & Arthur, M. A. *Soc. Econ. Paleont. Mineral. Spec. Publ.* **25**, 19–50 (1977).
9. Stehli, F. G., Douglas, R. G. & Newell, N. D. *Science* **164**, 947–949 (1969).
10. Hart, M. B. *Nature* **286**, 252–254 (1980).
11. Steineck, P. L. & Fleischer, R. L. *J. Paleont.* **52**, 618–635 (1978).
12. Saunders, J. B. *et al. Init. Rep. DSDP* Leg **15**, 796–771 (1973).
13. Bolli, H. M. *Init. Rep. DSDP* Leg **4**, 557–643 (1970).
14. Bolli, H. M. & Premoli Silva, I. *Init. Rep. DSDP* Leg **15**, 475–497 (1973).
15. Premoli Silva, I. & Bolli H. M. *Init. Rep. DSDP* Leg **15**, 499–547 (1973).
16. Blow, H. W. *Int. Conf. Plank. Microfossils*, Geneva, 199–422 (1969).
17. Bolli H. M. *Bull. U.S. Nat. Mus.* **215**, 97–123 (1957); **215**, 155–172 (1957).
18. Bolli, H. M. & Bermudez, P. J. *Bull. Ass. Venez. geol. Miner. Petrol.* **8**, 119–149 (1965).
19. Berggren, W. A. *Lethaia* **5**, 195–215 (1972).
20. Savin, S. M. *A. Rev. Earth planet. Sci.* **5**, 319–355 (1977).
21. Kennett, J. P. & Shackleton, N. J. *Nature* **260**, 513–515 (1976).
22. Shackleton, N. J. & Kennett, J. P. *Init. Rep. DSDP* Leg **29**, 743–755 (1975).
23. Berggren, W. A. *Init. Rep. DSDP* Leg **12**, 953–963 (1972).
24. Shackleton, N. J. & Kennett, J. P. *Init. Rep. DSDP* Leg **29**, 801–807 (1975).
25. Shackleton, N. J. & Opdyke, N. D. *Nature* **270**, 216–217 (1977).
26. Keigwin, L. D. & Thunell, R. C. *Nature* **282**, 294–296 (1979).
27. Berggren, W. A. *Proc. 11th Plank. Conf.*, Rome, 41–56 (1971).
28. Boersma, A. & Shackleton, N. J. *Init. Rep. DSDP* Leg, **39**, 911–924 (1977).
29. Vergnaud-Grazzini, C. *Init. Rep. DSDP* Leg **47**, 507–511 (1979).
30. Corliss, B. H. *Nature* **282**, 63–65 (1979).
31. Vail, P. R., Mitchum, R. M. Jr & Thompson, S. *Mem. Am. Ass. petrol. Geol.* **26**, 83–97 (1977).

Rates of emission of H_2S from plants and patterns of stable sulphur isotope fractionation

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In all green plants both HSO_3^- and SO_4^{2-} are reduced during photosynthesis, and many species can emit the product as H_2S gas. Plants will emit H_2S after fumigation with acute doses of SO_2 gas[1,2], irrigation with 5% K_2SO_4 solutions[3], or immersion of roots in HSO_3^- and SO_4^{2-} solutions[4]. If such emissions of H_2S by plants occur in nature, then this process adds sulphur to the atmosphere and contributes to the global sulphur budget. Oxidation of biogenic H_2S in the atmosphere may also contribute to the formation of acid rain. We report here that irradiated plants with roots immersed in HSO_3^- and SO_4^{2-} solutions at concentrations found in nature emit H_2S from their leaves, and that the fractionation of stable isotopes of sulphur during H_2S emission may be useful for identifying atmospheric sulphur that has been generated by photosynthetic reduction of sulphur.

Rates of emission of H_2S were measured for 10-week-old soybean plants (*Glycine max* L. Merr. cvs. Kent, Peking and York) raised in growth cabinets. Days were 12 h long, photosynthetically active radiation (PAR) was about 600 $\mu E\ m^{-2}\ s^{-1}$, humidity was 50–60% and the temperature was 15 °C at night and 20 °C during the day. Plants were prepared for experiments by removing them from potting media, washing and trimming their roots, and placing them in a flask with distilled water. The flask and plants were placed in a flow-through Plexiglass chamber, the upper portion of which was isolated by a diaphragm. Free space around the canopy was reduced by foam spacers inserted into the chamber, and air around the leaves was mixed by a propeller, driven by an electrical motor mounted outside the chamber. The roots were aerated by one airstream, and another airstream was passed through activated charcoal and used to flush the air space around the leaves. The chamber wall, diaphragm, foam spacers and propeller were lined with FEP Teflon to reduce H_2S absorption. After 30 min of incubation, the chamber was sealed, and air leaving it was analysed for H_2S with an Interscan Model 1170 sp H_2S analyser. Sampling was limited to 15 min to prevent water vapour from condensing in the chamber. Radiation by Sylvania Cool Lux bulbs was filtered through water and was 500 $\mu E\ m^{-2}\ s^{-1}$ (300–760 nM) inside the chamber.

Plants in distilled water were tested for H_2S emission, then Na_2SO_4 (Bäker, analytical grade) was added to the flask to increase the amounts of SO_4^{2-} available to the roots. Concentrations of SO_4^{2-} in the test solutions were 0.04, 0.1 and 1.0 mM. The highest concentration used is thought to be optimal for growth of agricultural crops[5], and the lowest concentration is similar to that of acid rain in the eastern US[6]. After treatment with the series of SO_4^{2-} solutions, plants were dried in an oven and rates of H_2S emission calculated on a plant dry-weight basis using the formula:

$$J_{H_2S} = XFW^{-1} \quad (1)$$

where J_{H_2S} is the H_2S emission rate ($ng\ g^{-1}\ s^{-1}$), X the H_2S concentration leaving the chamber ($ng\ cm^{-3}$), F the airstream flow rate ($cm^3\ s^{-1}$) and W plant dry weight (g).

0028–0836/81/080672—02$01.00

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A similar system was used to determine the fractionation of stable sulphur isotopes during emission. We used 4-week-old cucumber plants (*Cucumis sativus* L. cv. Straight Eight) grown in a greenhouse, because these were found to emit H_2S in previous experiments[4]. The isotopic composition of the test solutions (0.1–0.2 M HSO_3^- or SO_4^{2-}) was known. To obtain sufficient H_2S for analysis with a mass spectrometer, sulphur ions were at a higher concentration in test solutions for these experiments than for determining rates of emission. The H_2S emitted by the plants was trapped as Ag_2S by filtering the air leaving the chamber through $AgNO_3$ deposited on quartz wool. This procedure was repeated several times with fresh plants and fresh test solution until several mg of Ag_2S were collected. The H_2S traps showed no selectivity for sulphur isotopes. After these experiments, leaves were oven dried, and sulphur was combusted to SO_4^{2-} in a Parr bomb. Washings were treated with Ba^{2+} to precipitate $BaSO_4$ and then chemically reduced to yield Ag_2S[7]. $\delta^{34}S$ values (Table 1) for Ag_2S from both leaf sulphur and H_2S gas were determined by isotope ratio mass spectrometry by reacting Ag_2S with Cu_2O at 900 °C, to form SO_2.

H_2S was detected from plants in all test solutions containing SO_4^{2-} (Fig. 1). Even plants in the most dilute solutions (0.04 mM) had a mean emission rate of 0.036 ng g^{-1} s^{-1}. Rates of emission increased as the concentration of SO_4^{2-} in the test solution increased, and the highest rate recorded was 0.07 ng g^{-1} s^{-1} for plants treated with a test solution of 1.0 mM SO_4^{2-}. H_2S emission rates persisted for several hours when experiments were prolonged. Emission rates observed here are lower than the 10 μg g^{-1} s^{-1} observed for detached cucumber leaves treated with 25–100 mM SO_4^{2-} and higher than the 0.6 pg g^{-1} s^{-1} observed for spruce seedlings[3]. When the lights were turned off after two experiments, emission rates declined within 15 min to about 30% of the highest rate. Other workers have also observed the light dependence of SO_4^{2-} reduction[8,9] and H_2S emission[1,2,4]. No H_2S was emitted from plants placed in distilled water, and there seemed to be no consistent differences in emission rate between the three cultivars at any single concentration of SO_4^{2-}.

If SO_4^{2-} is available to plants at a concentration of 0.05 mM and they emit H_2S at a rate of 0.04 ng g^{-1} s^{-1}, then the amount of H_2S produced worldwide from photosynthetic reduction of SO_4^{2-} could be 5.4×10^{13} g yr^{-1}. This constitutes about 50% of the amount needed to balance the global sulphur budget[10,11]. Our calculations were made by estimating global biomass at 1.85×10^{12} dry tons[12], by assuming that about 5% of this weight is composed of leaves[13], and then by averaging the daily photoperiod to 12 h. Plants may emit H_2S at greater rates if SO_4^{2-} concentrations are generally greater than 0.05 mM. In addition, H_2S emitted by plants may be oxidized in the atmosphere and eventually returned to soils in the form of acid rain. A single sulphur atom could conceivably be cycled many times between the soil and atmosphere by the processes of H_2S emission from plants and acid rain.

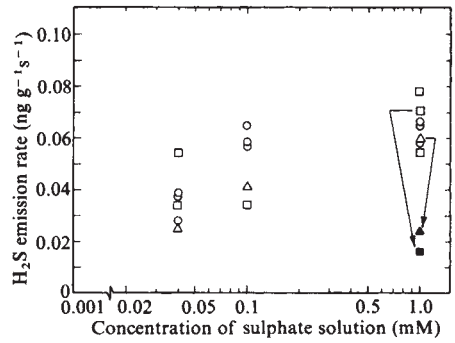

Fig. 1 H_2S emission rates (plant dry-weight basis) from irradiated soybean plants (open symbols) and from two plants transferred to darkness for a period of 15 min (closed symbols). Emission rates were monitored for three cultivars: ○, Kent; □, York; △, Peking.

H_2S emitted by plants contained more ^{32}S and less ^{34}S than either the leaves or the test solutions (Table 1). $\delta^{34}S$ values differed by −9.7‰ from the SO_4^{2-} source and ~−16.8‰ from the HSO_3^- source. The more extreme isotope fractionation pattern observed for HSO_3^- was also observed when both oxidized sulphur ions were supplied to plants in one solution. These results suggest that when both HSO_3^- and SO_4^{2-} are available, the emitted H_2S originates from the HSO_3^- source.

Photosynthetic sulphur-reducing bacteria show similar ^{32}S selectivity during H_2S formation[14]. Thus, depletion of ^{34}S resulting from isotope fractionation during H_2S formation may be common to chloroplasts and some prokaryotes. The ratio of isotopic rate constants for S–O bond rupture during inorganic reduction is similar ($k_{32}/k_{34}=1.022$)[15] to the fractionations seen in our experiments.

We suggest that stable sulphur isotope analysis may indicate whether or not atmospheric sulphur compounds have originated from vegetation. Atmospherics derived from H_2S emissions from plants should be enriched in ^{32}S compared with stable sulphur isotopes that are available to plant roots. These fractionation patterns are independent of the oxidation state of sulphur in the air. We have observed that the $\delta^{34}S$ values of plants reflect $\delta^{34}S$ values of sulphur in their environment[16–18], and we suggest that plants that emit H_2S should become enriched in ^{34}S relative to their sulphur source.

This research was supported by NRC (Canada) grants to J.D.B. and H.R.K. and a grant to H.A.M. from the Electrical Power Research Institute (USA). We thank Dr Peter Vitousek for commenting on an earlier draft of this manuscript.

Table 1 Sulphur isotope abundances in test solutions, cucumber leaves and emitted H_2S on exposure of plant roots to HSO_3^- and SO_4^{2-} solutions of different concentrations

	$\delta^{34}S$		
	Solution*	Leaves	H_2S
Control	—	+10.7	—
Treatment with:			
0.1 M HSO_3^-	+2.2	+4.5	−14.5
0.2 M HSO_3^-	+2.2	+2.7	−14.6
0.2 M SO_4^{2-}	+4.0	+3.1	−5.7
0.1 M HSO_3^- + 0.1 M SO_4^{2-}	+3.1	+3.9	−13.4

Using accepted practice, $\delta^{34}S$ values are defined with respect to meteoritic troilite as

$$\delta^{34}S=[^{34}S/^{32}S]_{sample}/[^{34}S/^{32}S]_{meteorite}-1\times10^3$$

Control; no H_2S was emitted from plants treated with distilled water.
* $\delta^{34}S$ value before experimentation with plants.

Received 29 September; accepted 3 December 1980.

1. Decormis, L. *C. r. hebd Séanc Acad. Sci.* D **266**, 683–685 (1968).
2. DeCormis, L. *Proc. 1st Eur. Congr. on the Influence of Air Pollution on Plants and Animals, Wageningen*, 75–78 (1969).
3. Spaleny, J. *Pl. Soil* **48**, 557–563 (1977).
4. Wilson, L. G., Bressan, R. A. & Filner, P. *Pl. Physiol.* **61**, 184–189 (1978).
5. Anderson, J. W. *Sulphur in Biology* Studies in Biology no. 101. (Arnold, London, 1978).
6. Likens, G. E. & Bormann, F. H. *Science* **184**, 1176–1179 (1974).
7. Thode, H. G., Monster, J. & Dunford, H. B. *Geochim. cosmochim. Acta* **25**, 159–174 (1961).
8. Schmidt, A. & Trebst, A. *Biochim. biophys. Acta* **180**, 529–535 (1969).
9. Trebst, A. & Schmidt, A. *Prog. Photosyn. Res.* **3**, 1510–1516 (1969).
10. Kellogg, W. W., Cadle, R. D., Allen, E. R., Lazrus, A. L. & Martell, E. A. *Science* **175**, 587–596 (1972).
11. Nguyen, B. D., Gaudry, A., Bonsang, B. & Lambert, G. *Nature* **275**, 637–639 (1978).
12. Whittaker, R. H. *Communities and Ecosystems*, 83 (Macmillan, New York, 1970).
13. Odum, E. P. *Fundamentals of Ecology*, 376 (Saunders, Philadelphia, 1971).
14. Kemp, A. L. W. & Thode, H. G. *Geochim. cosmochim. Acta* **32**, 71–91 (1958).
15. Harrison, A. G. & Thode, H. G. *Trans. Faraday Soc.* **53**, 1648–1651 (1957).
16. Krouse, H. R. *Nature* **265**, 45–46 (1977).
17. Winner, W. E., Bewley, J. D., Brown, H. M. & Krouse, H. R. *Oecologia* **36**, 351–361 (1978).
18. Case, J. W. & Krouse, H. R. *Oecologia* **44**, 248–257 (1980).

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The stability of real ecosystems

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Most current theoretical work on whole ecological communities is based on a conception of community dynamics (see, for example, refs 1, 2) in which the community resides in a neighbourhood of equilibrium. However, there is very little evidence that this is a realistic viewpoint. To test this fundamental assumption 'empirically', I have constructed here plausible community matrices[1,2] corresponding to 40 real food webs[3] and studied their local stability properties. I find that stability (in the sense of tending to return to the presumed equilibrium after a small perturbation) is far more likely if the interaction strengths are chosen in accord with the nature of the particular organisms in each food web, rather than strictly at random. Either this is a monumental coincidence, or the equilibrium viewpoint really is appropriate for quite a few real communities. Additional results of this analysis are that stability generally requires a degree of intraspecific interference on the part of some consumer species, whereas interspecific interference tends to exert a destabilizing influence.

As emphasized by Pimm and Lawton[4], element A_{ij} of the community matrix gives the *per capita* effect of species j on species i in a neighbourhood of the presumed equilibrium. This is an important observation, for it enables one to make plausible guesses about the relative strengths of some interactions. Thus, the *per capita* effect of an insect population on a tree population is much smaller than the *per capita* effect of trees on insects, and so on[4]. An additional advantage of this approach is that it treats the community matrix as a truly local object, which in reality it is. Thus, the results reported here are completely independent of any assumptions about global dynamics; in particular, these are not Lotka–Volterra models.

For any real food web, we can construct a plausible community matrix by specifying four real, positive parameters f_1, s_1, f_2, s_2, with f_1 and f_2 chosen from the interval [0, 1], and applying the following rules (which also serve to define the parameters f_1, s_1, f_2, s_2): (1) Trophic interactions. These matrix elements are chosen as by Pimm and Lawton[4] when their discussion applies. Cases not covered by Pimm and Lawton are treated in the spirit of their approach. It is in this step that a pattern of interaction strengths is arrived at which expresses the nature of the particular organisms in the food web. (2) Intraspecific interference. If species i is a primary producer, matrix element A_{ii} is chosen at random from the interval $(-s_1, 0)$. In addition, a fraction f_1 of the consumer species, chosen at random, is given diagonal matrix elements chosen at random from the interval $(-s_1, 0)$. The diagonal matrix elements for all other consumer species are set equal to zero. (3) Interspecific interference. If two consumers i and j have a prey species in common, then these species may indulge in interspecific interference, and I will call the ordered pair (i, j) 'potential competitors'. (Interspecific interference can also arise from other causes, such as competition for nest sites, but I will neglect such interactions here.) For a fraction f_2 of the potential competitors, chosen at random, the corresponding matrix elements A_{ij} are decreased from the values assigned in step (1) by amounts chosen at random from the interval $(0, s_2)$.

The food webs used in this study are the 40 webs recently compiled from the published literature by Briand[3], which is the most thorough existing community food web collection. For a variety of parameter sets $\{f_1, s_1, f_2, s_2\}$, 100 plausible community matrices were constructed for each Briand food web, using the above rules and a pseudorandom number generator, and the fraction of stable community matrices corresponding to each food web was computed. The results of some of these computations are displayed in Tables 1–4.

Table 1 shows the effect of increasing the strength s_1 of the intraspecific interference (relative to, of course, the strength of trophic interactions). In these computations, $f_1 = 1$ (all species in the community experience intraspecific interference) and $f_2 = 0$ (there is no interspecific interference). It is readily seen that stronger intraspecific interference favours stability, just as one expects.

Table 2 shows the effect of increasing the fraction f_1 of consumer species which engage in intraspecific interference. In these computations $f_2 = 0$ again, and $s_1 = 10^6$. Because of this large value for s_1 and in view of Tables 1 and 3, the values given in Table 2 can be viewed as upper bounds on the fraction of stable systems. It appears that at least one-tenth and perhaps as much as one-half of the consumer species in these communities need to exhibit some degree of intraspecific interference if the equilibrium viewpoint is to apply.

Pimm and Lawton[4] use the term 'self-limitation' for what I am calling intraspecific interference (namely, negative A_{ii}), and they

Table 1 Effect of changes in the strength of intraspecific competition on stability

Case no.	Community	Fraction of community matrices which are stable $s_1 = 0.1$	$s_1 = 1$	$s_1 = 10$	$s_1 = 100$
1	Cochin estuary	0.00	0.12	0.99	1.00
2	Krysna estuary	0.00	0.64	1.00	1.00
3	Long Island estuary	0.01	0.49	1.00	1.00
4	California salt marsh	0.00	0.52	1.00	1.00
5	Georgia salt marsh	1.00	1.00	1.00	1.00
6	California tidal flat	0.00	0.07	0.99	1.00
7	Narragansett Bay	0.00	0.00	0.93	1.00
8	Bissel Cove marsh	0.00	0.07	0.96	1.00
9	Lough Ine rapids	0.01	0.43	0.99	1.00
10	Exposed intertidal (New England)	0.41	0.89	1.00	1.00
11	Protected intertidal (New England)	0.13	0.59	1.00	1.00
12	Exposed intertidal (Washington State)	0.00	0.41	0.99	1.00
13	Protected intertidal (Washington State)	0.00	0.19	1.00	1.00
14	Mangrove swamp (Station 1)	0.02	0.20	0.98	1.00
15	Mangrove swamp (Station 2)	0.01	0.24	0.99	1.00
16	Pamlico River	0.01	0.45	1.00	1.00
17	Marshallese reefs	0.00	0.39	0.99	1.00
18	Kapingamarangi atoll	0.00	0.33	1.00	1.00
19	Moosehead Lake	0.01	0.74	1.00	1.00
20	Antarctic pack ice zone	0.00	0.09	0.95	1.00
21	Ross Sea	0.00	0.12	0.95	1.00
22	Bear Island	0.00	0.01	0.92	1.00
23	Canadian prairie	0.00	0.19	0.97	1.00
24	Canadian willow forest	0.07	0.92	1.00	1.00
25	Canadian aspen forest	0.12	0.95	1.00	1.00
26	Aspen parkland	0.00	0.28	0.98	1.00
27	Wytham wood	0.00	0.12	0.97	1.00
28	New Zealand salt-meadow	0.14	0.78	1.00	1.00
29	Arctic seas	0.00	0.07	0.97	1.00
30	Antarctic seas	0.00	0.47	1.00	1.00
31	Black Sea epiplankton	0.00	0.00	0.82	1.00
32	Black Sea bathyplankton	0.00	0.00	0.76	1.00
33	Crocodile Creek	0.01	0.87	0.99	1.00
34	River Clydach	0.00	0.07	0.97	1.00
35	Morgan's Creek	0.00	0.71	1.00	1.00
36	Mangrove swamp (Station 6)	0.00	0.26	1.00	1.00
37	California sublittoral	0.01	0.76	1.00	1.00
38	Lake Nyasa rocky shore	0.00	0.34	1.00	1.00
39	Lake Nyasa sandy shore	0.00	0.63	1.00	1.00
40	Malaysian rain forest	0.00	0.67	1.00	1.00

0028–0836/81/080674—03$01.00

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Table 2 Effect of changes in the frequency of intraspecific competition on stability

Case no.	Community	Fraction of community matrices which are stable $f_1=0.0$	$f_1=0.1$	$f_1=0.5$	$f_1=1.0$
1	Cochin estuary	0.00	0.00	0.51	1.00
2	Krysna estuary	0.00	0.05	0.60	1.00
3	Long Island estuary	0.00	0.03	0.56	1.00
4	California salt marsh	0.00	0.04	0.62	1.00
5	Georgia salt marsh	1.00	1.00	1.00	1.00
6	California tidal flat	0.00	0.01	0.42	1.00
7	Narragansett Bay	0.00	0.00	0.32	1.00
8	Bissel Cove marsh	0.00	0.02	0.53	1.00
9	Lough Ine rapids	0.00	0.01	0.44	1.00
10	Exposed intertidal (New England)	0.53	0.61	0.89	1.00
11	Protected intertidal (New England)	0.15	0.16	0.72	1.00
12	Exposed intertidal (Washington State)	0.00	0.00	0.27	1.00
13	Protected intertidal (Washington State)	0.00	0.00	0.29	1.00
14	Mangrove swamp (Station 1)	0.11	0.34	0.85	1.00
15	Mangrove swamp (Station 2)	0.02	0.16	0.61	1.00
16	Pamlico River	0.21	0.32	0.79	1.00
17	Marshallese reefs	0.00	0.00	0.38	1.00
18	Kapingamarangi atoll	0.00	0.02	0.51	1.00
19	Moosehead Lake	0.00	0.01	0.41	1.00
20	Antarctic pack ice zone	0.00	0.01	0.44	1.00
21	Ross Sea	0.00	0.00	0.38	1.00
22	Bear Island	0.00	0.00	0.21	1.00
23	Canadian prairie	0.00	0.02	0.44	1.00
24	Canadian willow forest	0.00	0.10	0.65	1.00
25	Canadian aspen forest	0.00	0.00	0.11	1.00
26	Aspen parkland	0.00	0.00	0.21	1.00
27	Wytham wood	0.00	0.02	0.50	1.00
28	New Zealand salt-meadow	0.00	0.00	0.05	1.00
29	Arctic seas	0.00	0.00	0.13	1.00
30	Antarctic seas	0.00	0.00	0.23	1.00
31	Black Sea epiplankton	0.00	0.00	0.12	1.00
32	Black Sea bathyplankton	0.00	0.00	0.08	1.00
33	Crocodile Creek	0.00	0.00	0.34	1.00
34	River Clydach	0.00	0.00	0.55	1.00
35	Morgan's Creek	0.00	0.00	0.34	1.00
36	Mangrove swamp (Station 6)	0.00	0.02	0.60	1.00
37	California sublittoral	0.00	0.00	0.34	1.00
38	Lake Nyasa rocky shore	0.00	0.00	0.10	1.00
39	Lake Nyasa sandy shore	0.00	0.00	0.07	1.00
40	Malaysian rain forest	0.00	0.27	0.82	1.00

Table 3 Effect of interspecific competition on stability

Case no.	Community	Fraction of community matrices which are stable $f_2=0.0$	$f_2=0.1$	$f_2=0.5$	$f_2=1.0$
1	Cochin estuary	0.12	0.10	0.09	0.03
2	Krysna estuary	0.64	0.60	0.60	0.72
3	Long Island estuary	0.49	0.39	0.41	0.33
4	California salt marsh	0.52	0.46	0.37	0.18
5	Georgia salt marsh	1.00	1.00	1.00	1.00
6	California tidal flat	0.07	0.04	0.02	0.01
7	Narragansett Bay	0.00	0.00	0.00	0.00
8	Bissel Cove marsh	0.07	0.04	0.02	0.03
9	Lough Ine rapids	0.43	0.42	0.40	0.35
10	Exposed intertidal (New England)	0.89	0.93	0.91	0.91
11	Protected intertidal (New England)	0.59	0.55	0.49	0.60
12	Exposed intertidal (Washington State)	0.41	0.36	0.35	0.42
13	Protected intertidal (Washington State)	0.19	0.28	0.48	0.35
14	Mangrove swamp (Station 1)	0.20	0.26	0.30	0.29
15	Mangrove swamp (Station 2)	0.24	0.18	0.24	0.26
16	Pamlico River	0.45	0.40	0.34	0.28
17	Marshallese reefs	0.39	0.39	0.27	0.19
18	Kapingamarangi atoll	0.33	0.28	0.01	0.00
19	Moosehead Lake	0.74	0.73	0.48	0.35
20	Antarctic pack ice zone	0.09	0.04	0.02	0.02
21	Ross Sea	0.12	0.06	0.00	0.01
22	Bear Island	0.01	0.00	0.00	0.00
23	Canadian prairie	0.19	0.17	0.09	0.04
24	Canadian willow forest	0.92	0.86	0.60	0.33
25	Canadian aspen forest	0.95	0.84	0.54	0.43
26	Aspen parkland	0.28	0.21	0.04	0.00
27	Wytham wood	0.12	0.08	0.03	0.00
28	New Zealand salt-meadow	0.78	0.58	0.15	0.03
29	Arctic seas	0.07	0.08	0.03	0.02
30	Antarctic seas	0.47	0.32	0.07	0.02
31	Black Sea epiplankton	0.00	0.00	0.00	0.00
32	Black Sea bathyplankton	0.00	0.00	0.00	0.00
33	Crocodile Creek	0.87	0.54	0.07	0.00
34	River Clydach	0.07	0.08	0.03	0.04
35	Morgan's Creek	0.71	0.72	0.38	0.29
36	Mangrove swamp (Station 6)	0.26	0.27	0.27	0.22
37	California sublittoral	0.76	0.69	0.32	0.15
38	Lake Nyasa rocky shore	0.63	0.23	0.00	0.00
39	Lake Nyasa sandy shore	0.67	0.49	0.18	0.07
40	Malaysian rain forest	0.34	0.35	0.12	0.00

argue[5] that it rarely occurs for consumer species. They seem to feel that negative A_{ii} means that intraspecific interference is not only present, but is furthermore the 'limiting factor' in the sense of Liebig. But surely any intraspecific interference, whether it is 'limiting' or not, will manifest itself in the community matrix as negative A_{ii}. For this reason I have avoided Pimm and Lawton's terminology, and feel that negative A_{ii} can be expected to occur quite often for consumer species.

The effect of interspecific interference is investigated in Table 3, with the parameter values $f_1=1$, $s_1=1$, $s_2=0.1$ fixed, while f_2, the fraction of potential competitors which participate in interspecific interference, varies. It can be seen that these interactions usually exert a destabilizing influence.

Finally, Table 4 explores the plausibility of the equilibrium viewpoint itself. The first column of this table gives the fraction of stable community matrices, constructed as above with $\{f_1, s_1, f_2, s_2\}=\{1, 1, 0, 0\}$. The second column was obtained as follows: after constructing each community matrix as above, all the positive and all the negative off-diagonal entries were randomly permuted, and then stability was checked. Thus, all predator–prey relationships were preserved, as was the average interaction strength in each matrix, but the detailed pattern of interaction strengths suggested by the particular organisms in each food web (step (1) above) was destroyed. Clearly, local stability was destroyed along with this pattern.

The third column in Table 4 results from a somewhat weaker disruption of the pattern of interaction strengths. In this case, for each community matrix constructed according to the original rules, the pairs of non-zero elements (A_{ij}, A_{ji}) were randomly permuted. Again, local stability is destroyed.

If the equilibrium viewpoint is not appropriate for these communities, there is no reason for local stability of the community matrix to be associated with a more plausible pattern of interaction strengths.

Other evidence in favour of the equilibrium viewpoint, though in a somewhat different conceptual framework from that embodied by ref. 1, has come from some simulation studies[6]. In a study which is closely related to the present one in spirit (published shortly after the submission of this paper), Lawlor[7] has found, for 11 avian and 10 reptilian competitive guilds, that

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Table 4 Effect of trophic interaction pattern on stability

Case no.	Community	Fraction of community matrices which are stable 'Normal'	Disruption 1	Disruption 2
1	Cochin estuary	0.12	0.05	0.00
2	Krysna estuary	0.64	0.00	0.00
3	Long Island estuary	0.49	0.00	0.00
4	California salt marsh	0.52	0.03	0.00
5	Georgia salt marsh	1.00	0.22	1.00
6	California tidal flat	0.07	0.00	0.00
7	Narragansett Bay	0.00	0.00	0.00
8	Bissel Cove marsh	0.07	0.02	0.00
9	Lough Ine rapids	0.43	0.03	0.12
10	Exposed intertidal (New England)	0.89	0.15	0.50
11	Protected intertidal (New England)	0.59	0.12	0.16
12	Exposed intertidal (Washington State)	0.41	0.00	0.00
13	Protected intertidal (Washington State)	0.19	0.00	0.00
14	Mangrove swamp (Station 1)	0.20	0.13	0.15
15	Mangrove swamp (Station 2)	0.24	0.09	0.16
16	Pamlico River	0.45	0.00	0.12
17	Marshallese reefs	0.39	0.00	0.00
18	Kapingamarangi atoll	0.33	0.00	0.00
19	Moosehead Lake	0.74	0.00	0.00
20	Antarctic pack ice zone	0.09	0.00	0.02
21	Ross Sea	0.12	0.00	0.00
22	Bear Island	0.01	0.00	0.00
23	Canadian prairie	0.19	0.00	0.00
24	Canadian willow forest	0.92	0.04	0.02
25	Canadian aspen forest	0.95	0.02	0.04
26	Aspen parkland	0.28	0.00	0.00
27	Wytham wood	0.12	0.00	0.00
28	New Zealand salt-meadow	0.78	0.00	0.13
29	Arctic seas	0.07	0.00	0.02
30	Antarctic seas	0.47	0.00	0.00
31	Black Sea epiplankton	0.00	0.00	0.00
32	Black Sea bathyplankton	0.00	0.00	0.00
33	Crocodile Creek	0.87	0.00	0.00
34	River Clydach	0.07	0.00	0.00
35	Morgan's Creek	0.71	0.00	0.00
36	Mangrove swamp (Station 6)	0.26	0.00	0.00
37	California sublittoral	0.76	0.00	0.00
38	Lake Nyasa rocky shore	0.63	0.00	0.00
39	Lake Nyasa sandy shore	0.67	0.00	0.00
40	Malaysian rain forest	0.34	0.02	0.11

randomizations of community matrices constructed from measured resource overlaps often have a deleterious effect on stability. Although this result is suggestive, its bearing, like that of single-species studies[8], on the stability and dynamics of whole communities is not entirely clear.

Auerbach[9], applying Pimm and Lawton's community matrix rules to certain host-parasitoid systems, did not find any stable matrices. This is surely due in large part to the fact that he did not extend Pimm and Lawton's rules, as I have, to allow for varying degrees of intra- and interspecific interference. As pointed out by several authors[10–12], and reinforced by Tables 1 and 2, community matrix stability depends crucially on the extent of intraspecific interference (both in commonness of occurrence and in strength).

It has long been known that solutions to systems of autonomous ordinary differential equations can behave very strangely ('recurrent trajectories')[13,14]. More recently, the realization has grown that even very simple systems can exhibit very complicated dynamical behaviour ('chaos')[15,16]. For ecosystems, biology provides additional complications, such as seasonality, spatial migration, invasion, local extinction, fluctuating extrinsic factors, catastrophic extrinsic factors and phenotypic plasticity. In the face of all these complications, the equilibrium viewpoint is hardly to be taken for granted.

The above findings in favour of the equilibrium viewpoint legitimize a large body of ecological theory and suggest that it is worth pursuing this viewpoint further. At the same time, we should be aware of the many factors, both mathematical and biological, which complicate the picture, and strive to incorporate them into our ecological world-view.

I thank Frédéric Briand for providing me with his collection of food webs in advance of publication, and for some most interesting discussions. I also thank the University of Guelph for the provision of a substantial amount of computer time under its Special Computing Program.

Received 8 September; accepted 2 December 1980.

1. May, R. M. *Stability and Complexity in Model Ecosystems* (Princeton University Press, 1974).
2. Levins, R. in *Ecology and Evolution of Communities* (eds Cody, M. L. & Diamond, J. M.) 16–50 (Harvard University Press, 1975).
3. Briand, F. *Am. Nat.* (submitted).
4. Pimm, S. L. & Lawton, J. H. *Nature* **275**, 542–544 (1978).
5. Lawton, J. H. & Pimm, S. L. *Nature* **272**, 189–190 (1978).
6. Hall, C. A. S. & Day, J. W. (eds) *Ecosystem Modelling in Theory and Practice* (Wiley, New York, 1977).
7. Lawlor, L. R. *Am. Nat.* **116**, 394–408 (1980).
8. Hassell, M. P. *J. Anim. Ecol.* **49**, 603–628 (1980).
9. Auerbach, M. J. *Nature* **279**, 821–822 (1979).
10. Saunders, P. T. *Nature* **272**, 189 (1978).
11. Pimm, S. L. *Nature* **285**, 591 (1980).
12. Rejmánek, M. & Starý, P. *Nature* **285**, 591–592 (1980).
13. Birkhoff, G. D. *Dynamical Systems* (American Mathematical Society, New York, 1927).
14. Nemytskii, V. V. & Stepanov, V. V. *Qualitative Theory of Differential Equations* (Princeton Universiy Press, 1960).
15. Lorenz, E. N. *J. atmos. Sci.* **20**, 130–141 (1963).
16. Rössler, O. E. *Z. Naturforsch.* **31a**, 259–264, 1664–1670 (1976).

Perception of sign language from an array of 27 moving spots

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Many deaf people in the USA communicate in American sign language (ASL), which has an expressive capacity equivalent to that of a spoken language, although structurally independent of spoken languages[1,2]. It comprises hand and arm movements often combined with particular facial gestures; together these are sufficiently precise to transmit all the complexities and innuendoes of a language. Here we demonstrate that fluent ASL users can communicate easily when all they see of each other is an array of 27 light spots strategically placed on the hands and face. The results indicate the salient locations in normal sign perception, and suggest that it is feasible to transmit signs using the bandwidth of one telephone line rather than a much more expensive TV line.

In normal conditions, the understanding of sign language requires seeing the area in front of the signer's body from the top of the head to waist level, and within a few inches of each side—signs produced within a 6-inch radius of the chin are most precisely articulated[3]. Signs are differentiated from one another by hand shape (for example, index finger compared with little finger raised), hand orientation (for example, palm exposed rather than back of the hand exposed), type of movement (for example, movement towards compared with movement away from the signer), and 'place of articulation' (for example, a touch on the chin compared with a touch on the lips)[4,5]. Battison[5] estimated that there are 45 distinct hand shapes, 10 hand orientations, 10 movements and 25 locations—this indicates the precision required to distinguish signs. In addition it has been

0028–0836/81/080676—03$01.00

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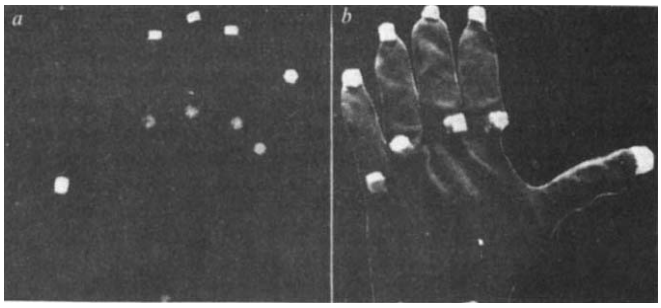

Fig. 1 A display of the back of a glove in *a*, lighting conditions in which only the reflections from the tape are visible and *b*, in normal lighting conditions.

suggested that facial expression may transmit critical information, perhaps redundantly[5], and that head movements may be criticial for differentiating affirmatives, negatives and interrogatives[6].

To determine which information is critical for evoking the perception of the various features, we adapted the method of Johansson *et al.*[7,8] to study grosser types of biological motion. Johansson attached patches of retroreflective tape to the principal joints of subjects—wrist, elbow, shoulder, ankle, knee and hip—then filmed them moving in special lighting conditions so that only the patches of tape were visible. The two-dimensional display of light spots produced was clearly and immediately identified by observers as human motion: a person walking, running, cycling, and such. Presentation of a still picture alone from the display resulted in perception of lights—the motion was necessary to convey the structural relation between the lights and thus to evoke perception of human action.

As signing comprises human movement, although faster, more finely detailed, and less repetitive than that studied by Johansson, it seemed possible that the same perceptual mechanisms which organized the spots of light in Johansson's displays into meaningful patterns could organize strategically placed light spots on the hands into hand shapes and motions. The problem was to derive a configuration of spots which would convey each of the important feature values in sign without confusion, and which would be immediately perceived as moving hands, suggesting that these points are normally the salient ones for sign perception. (Note that Poizner *et al.*[9] used a similar technique to code the motion patterns used for various inflections in ASL. For this purpose one light on the index fingertip and reference points are sufficient to distinguish between 50 movement types.)

We devised a glove with 13 strategically placed pieces of tape, the largest and brightest at the regions which convey most information: a large piece on the thumb tip and slightly smaller ones on each of the remaining fingertips. On the back of the hand there was a smaller spot on each finger, just proximal to the second joint from the tip and there were four even smaller spots around the wrist. The backs of two gloves are shown in Fig. 1 in lighting which shows only the retroreflective tape spots (*a*), and in normal lighting (*b*).

With the palm of the hand forward, an outline of the hand is perceived (from the fingertips and wrist boundary). There is no hand shape which can be assumed with the palm forward in which the fingertips are not visible—closing of the hand is evident from the motion and the approach of the tip spots towards the wrist. Elevation of individual fingers is clear from the position of the raised fingers relative to the folded ones.

The back of the hand appears as fingertips followed by two more rows of spots. The row at the joints is necessary because with the back of the hand forward it is possible to curve the fingers so that the tips are not visible, as in a fist. In this case, curvature of the fingers is apparent as a disappearance of the fingertip spots, but information on hand shape is still preserved by the relative positions of the joint spots.

Information about hand orientation is given again by motion and the configuration of spots. Each spot is actually a spherical protrusion which is therefore uniformly visible from a wide range of viewing angles. Motion information is readily apparent in horizontal and vertical directions, as it was for subjects used in other studies[8,9], from the changes in position of the spots. Motion in depth is conveyed by a change in the size and spacing of the spots due to foreshortening.

To convey place of articulation only one additional reference point was necessary—a spot on the nose. A touch at the eyebrows is seen as a hand motion above the middle of the face; at the shoulders, as outside and below the middle of the face. No further reference points on the body were necessary, perhaps because of the grosser articulation outside the face area. The spot on the nose, in addition to providing a reference point, also gave information on head movements (nodding, shaking and

S_1	S_2
	How is your work?
[confused, so shadows what saw] How is . . .	
	How is your work?
[now understands] OK, OK.	
	Same, same.
Same, same.	Boring
OK	
	Thinking new job, I mean transfer to another job? Still waiting for another job?
For you?	You. You!
me.	
Thinking about transferring to another job maybe.	
	Same place or other place?
Same. Here. But change job.	
	For how long have you been waiting?
No. No. No. Been thinking for a pretty long time to get transferred so I have to wait for my supervisor to help me get another job.	
	Are you going to night school this fall?
I'm thinking of writing to Middlesex County College for night college but I have to wait for the reply. Still waiting for a man who is responsible for picking the deaf people to put into one class. People.	
	It seems to me that you have been thinking for a very long time. You'd better stop thinking and do it now, before it becomes too late. OK? [Pauses with no response] Do you understand me?
Yes. Talk. That I'm not worried about it.	
	Noooo . . ! You have been concerned for a *long* time. Concerned. Not important to think. Do it before it becomes too late.
Oh. I understand. You mean that if I . . .	
	You wait too long. No don't. Waiting is not that good.
I understand.	
Today it's very hard to get into college. Very difficult. Because too many people want enrolment.	No. No.
	No. Do you know what law number 504 means to you? Always assume that it's not hard to get in because you are deaf. No. Enrol (any) time you want. 504 guarantees enrolment.
[S gives feedback]	
Yes. Yes. I get it. I understand.	
But how can I get in? I mean I have to get permission from someone to get in.	
	Get. You want to get into college you have to go to admissions office and ask what courses you want; then he will put down (what) you ask him. You pay an interpreter, you pay for, and if college says no, you bring 504 law and show them, and if college won't believe you go to a court and ask for some -- [interrupts herself and restarts immediately with] because I learned from my friend who refused - (I mean) college refused to pay nothing so she went to court and the lawyer went to the college and college realized their mistake and they will pay for all interpreters. (It's) easy. You don't have to pay nothing.
Oh. I understand.	[Pauses briefly, then teases with] Chicken, chicken.

Fig. 2 One conversation of 4 min transcribed into English from videotaped record. Subjects at this point had 10 min experience with the system. Average signing rate was ~80 signs per min. S_1, S_2, subjects 1 and 2 respectively.

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leaning forward), and provided feedback information to the person signing that the viewer was still watching.

Preliminary tests showed that effective communication at near normal rates is possible when viewing only the 27 points of light. Two pairs of deaf subjects were able to converse when viewing only the other's spots.Three of the four subjects were profoundly deaf, with hearing losses of 90 dB in the better ear. The fourth subject (C.F.) had some hearing and quite good speech. She and her partner were well educated with good reading and writing skills. Both learned ASL when young, and sign either ASL or English. The other two subjects had poor reading and writing skills and were only able to sign ASL. The transcript shown in Fig. 2 is an extract from the conversation of the first pair, translated into 'English' by C.F.

Each member of the pair was seated in a private room ~3 feet from a 21-inch TV monitor and a camera. The gloves and nose spot were placed on the subject, who was allowed a few minutes of self-viewing in normal lighting conditions. Then all the lights in each room except the one next to the TV camera were turned out, and the brightness and contrast of the TV monitors adjusted so that each subject could see only the spots. (Note that the camera was set so that movement within the signing space would be visible; thus the screen image of the spots was smaller than that displayed in Fig. 1. In addition, with movement, the spots commonly smear, leaving a slightly blurred image on each frame.) After self-viewing, the equipment was reconnected so that the subjects could see each other. They were instructed to discuss anything they wished and a free-flowing 20-min conversation ensued. Various topics were discussed by both pairs, with difficulties and requests for repetition only in finger spelling. A transcription of a portion of one pair's discussion is shown in Fig. 2. During this displayed segment the participants became quite heated—emotion was conveyed by the slow and abrupt movements of the spots. Nonmeaningful movements (for example, tugging at the gloves) were occasionally mimicked playfully, but no attempt was made to interpret them. Note that during debriefing both pairs of subjects reported that they really needed to concentrate, but that their conversation and understanding were normal except for finger spelling. Furthermore, all four subjects expressed their enjoyment of the system.

We believe that the picture that these subjects used could be coded to the capacity of one telephone line. This assumes that the useful information in each spot is its brightness or size, and position on the screen in time. A more controlled test is planned, using computer processing to ensure that this information alone is available.

We have thus shown that communication in sign language is possible using a highly stylized representation of the hands and body, at a considerably reduced information rate from a face-to-face situation. This demonstration suggests the possibility of developing a sign language telephone.

We acknowledge the assistance of Jay Jackson, Carol Furino, Cynthia Jefferey, Michael Androsko and Gene Marshall in testing our system. We also thank Lilah Axelsen for teaching us sign language, Eric Albert and Carol Furino for help in interpreting tapes, Carol Tipton and Susan Fischer for consultation about deaf communication and sign language, and Bela Julesz and Osamu Fujimura for helpful suggestions. A portion of this paper was presented at the Nato Advanced Study Institute: Sign Language Research, in Copenhagen, August 1979.

Received 22 July; accepted 18 December 1980.

1. Bellugi, U. & Klima, E. S. in *Origin and Evolution of Language and Speech* (eds Harnad, S. R., Steklis, H. D. & Lancaster, J.) 515–538 (N.Y. Acad. Sci., 1976).
2. Grosjean, F. in *Recent Perspectives in American Sign Language* (eds Lane, H. & Grosjean, F.) (Lawrence Erlbaum, New Jersey, in the press).
3. Siple, P. in *Understanding Sign Language through Sign Language Research* 3–23 (Academic, New York, 1978).
4. Stokoe, W. C., Casterline, D. & Croneburg, C. *A Dictionary of American Sign Language on Linguistic Principles* (Gallaudet College Press, Washington DC, 1965).
5. Battison, R. *Lexical Borrowing in American Sign Language* (Linstok, Maryland, 1978).
6. Bellugi, U. & Fischer, S. *Cognition* **1**, 173–200 (1972).
7. Johansson, G. *Percept. Psychophys.* **14**, 201–211 (1973).
8. Johansson, G., von Hofsten, C. Jr & Jansson, G. *A. Rev. Psychol.* 27–63 (1980).
9. Poizner, H., Bellugi, U. & Lutes-Driscoll, V. *Journal of Experimental Psychology: Human Perception and Performance* (in the press).

Simultaneous yet independent inheritance of somatically acquired tolerance to two distinct H–2 antigenic haplotype determinants in mice

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We have previously presented evidence that somatically acquired immunological tolerance to foreign histocompatibility antigens induced in male inbred mice can be transmitted at a high frequency to first- and second-generation offspring without exposure of the progeny to the tolerizing antigenic stimulus[1]. These data have in turn been interpreted to support a hypothesis predicting soma to germ-line inheritance for acquired states of the immune system[2]. Formal proof of this genetic scheme at the molecular level is not yet available. However, an even stronger reason for abandoning the notion of the isolation of the germ line from the soma (Weismann's doctrine[2]) would be provided by a clear demonstration of the multiple inheritance of independently acquired somatic characters. Here we present evidence that individual male mice (B10) made tolerant to two different H–2 haplotypes (B10.BR, B10.D$_2$) can transmit the tolerance state to each haplotype independently, yet often simultaneously, at a high frequency to both first and second generation progeny.

Congeneic resistant mouse lines (B10.SgN, H–$2K^bD^b$; B10.D$_2$, H–$2K^dD^d$; B10.BR, H–$2K^kD^k$) were purchased at 6 weeks of age and ATH mice (H–$2K^sD^d$) at 8 weeks of age from Jackson Laboratories. The latter mice were used within 4 weeks of arrival. Normal litters of B10.SgN (hereafter called B10) were produced in our animal room from the Jackson foundation stock; some members of these litters were mated (brother × sister) to produce normal B10 animals on a regular basis during the 10-month study. F_1 hybrids (B10 × B10.D$_2$, B10 × B10.BR) were produced by matings from the Jackson foundation stock. These served as a source of lymphoid cells both for inducing and maintaining tolerance in the B10 mice, and as normal responder or stimulator cells in the cytotoxic T-cell assays.

Tolerance was induced in neonatal B10 mice (produced at the start of the breeding programme) by intraperitoneal (i.p.) inoculation of 100×10^6 F_1 lymphoid cells (approximately 1:1 mixtures of bone marrow and spleen cells, Fig. 1; bone marrow only, Fig. 2). As previously reported, repeated injections of the same type of F_1 cells were given at 2-week intervals throughout the study (i.p. injections until 4 weeks of age—thereafter intravenous injections were given). While certainly not essential to produce operational tolerance in such neonatally challenged mice[3,4], we have continued these regular inoculations of F_1 hybrid cells, as discussed earlier, to ensure high levels of circulating F_1 hybrid cells in the neonates. No data are yet available to judge whether such repeated challenge is essential to induce the transmission of tolerance from parents to offspring which we have shown subsequently occurs (ref. 1 and below).

Tolerant male mice were chosen for mating with normal B10 females beginning at 8 weeks of age. Tolerance in these breeding fathers has been assessed by both cell-mediated lympholysis (CML) responsiveness (performed after partial splenectomy under ether anaesthesia) and skin graft rejection (see legends to Figs 1, 2). Only animals judged to be tolerant by both criteria

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have been used in the breeding studies reported. However, responsiveness in the progeny of such animals has been assessed initially only by *in vitro* assay (see below). Comparison of skin grafting and CML responses of the progeny is being investigated. First-generation progeny of these animals were reared without exposure to F_1 antigens. At 8 weeks, spleen cells of individual progeny mice were assayed *in vitro* for their ability to give rise to cytotoxic lymphocytes after stimulation with irradiated (B10×B10.BR)F_1, (B10×B10.D_2)F_1, or third-party ATH (H–$2K^sD^d$) spleen cells (CML assay). Target cells in the ^{51}Cr-release test were concanavalin A blasts of the respective spleen cells used as stimulators[4–6].

Typical data for first-generation progeny of B10 male mice made tolerant to B10.BR or B10.D_2 determinants are shown in Fig. 1. Clearly these first-generation animals show a high frequency (some 40–50%) of tolerance (0–5% specific cytotoxicity) to those determinants used to induce tolerance in their fathers (B10.BR, Fig. 1 *d*; B10.D_2, Fig. 1*e*). Note that not only is the ability of the progeny to respond to the third-party antigens (ATH) unaffected but so too is their ability to respond to B10.D_2 antigens (progeny of males tolerant to B10.BR) or B10.BR antigens (progeny of males tolerant of B10.D_2). It is also clear (Fig. 1*f*) that first-generation mice born to males undergoing the tolerance-inducing treatment procedure, but which failed to become tolerant, are phenotypically (in terms of CML responses) equivalent to the offspring of normal B10 mice. These results confirm our earlier findings on the genetic transmission of acquired tolerance to A-strain H–2 antigens in CBA mice[1], and extend the generality of the phenomenon to two additional mouse strain combinations.

Clearly acquired tolerance to B10.D_2 antigens can be inherited independently of tolerance to B10.BR antigens (Fig. 1). Interestingly, an apparent simultaneous transmission of tolerance to the independent K/D-end specificities associated with a given haplotype (for example, H–2^k) has occurred, as these specificities are themselves independently expressed on separate molecules. Although this in itself may represent the simultaneous transmission of tolerance to independent antigenic determinants, note that responsiveness to the distinct K/D specificities was not tested independently in the recipients described here, and alloreactive cytotoxicity directed to K end-encoded determinants is more readily demonstrated than to D end-encoded determinants. Thus one interpretation of our data would be that responsiveness to major stimulating antigen determinants (K end-encoded) only was recorded in the CML assays used and there is no need to postulate the parallel transmission of immunological tolerance to K^k, D^k in animals bred from males tolerant to B10.BR cells. Indeed, recent studies (unpublished) indicate the independent transmission of tolerance to K^k, D^d from B10 male mice made tolerant to B10.A lymphoid tissue and subsequently mated with normal B10 female mice.

We have investigated whether tolerance to two different specificities can be independently transmitted to progeny using a situation in which individual B10 males were rendered tolerant simultaneously to both H–2^k and H–2^d haplotypes. Hereafter we will make the minimum interpretation that we are measuring unresponsiveness to the major stimulating specificities (K^k, K^d) associated with those haplotypes. The data in Fig. 2 summarize results from first-generation progeny (of four such males) obtained over a 12-week breeding period. The data are arranged as response profiles to B10.D_2 or ATH antigens for progeny mice grouped into different levels of responsiveness (% specific cytotoxicity) to B10.BR antigens. (A similar plot is obtained if the progeny are grouped into different levels of response to B10.D_2.) Note: (1) All progeny mice respond to ATH antigens, irrespective of their response patterns to each of the other two antigens tested. (2) The response pattern to one haplotype is not predictive of the level of responsiveness obtained towards the other haplotype (for example, of 80 progeny hyporesponsive to B10.BR—<10% cytotoxicity—40 respond in the normal range to B10.D_2 and 40 are tolerant or low responders to B10.D_2). That is, in contrast to their fathers which are tolerant of both haplotypes, many of these first-generation offspring are now tolerant for either one or the other haplotype. (3) Many of these progeny also score tolerant (27/150) to or are low responders (13/150) of both H–2 antigenic types. These results therefore show that the inheritance of

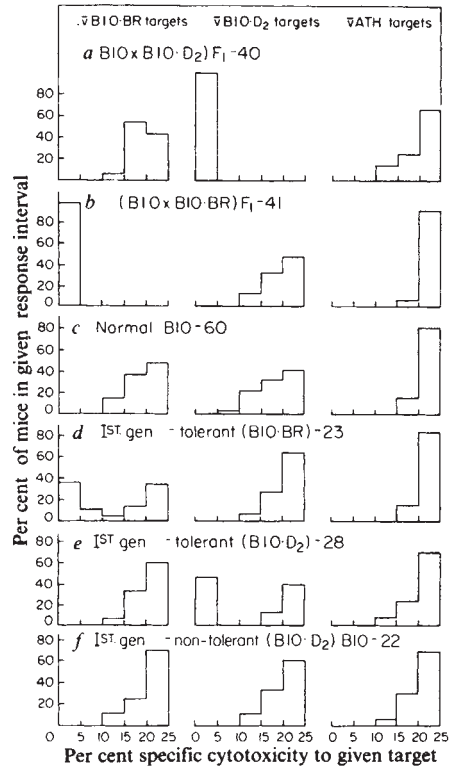

Fig. 1 CML response profiles of first-generation offspring of male B10 mice rendered tolerant to B10.BR or B10.D_2 antigenic determinants. Spleen cells (1.5×10^6 in duplicate) from individual 8-week progeny mice or control mice were stimulated in culture with irradiated (1,500 rad) spleen cells (5×10^5) of the relevant targets shown. Cytotoxicity was assayed at 5 days (refs 4–7). *a, b, c*, Responses of normal control mice (B10×B10.D_2)F_1, (B10×B10.BR)F_1 and B10 respectively. (The number of mice tested is indicated.) *d,e*, First-generation progeny of male B10, rendered neonatally tolerant to (B10×B10.BR)F_1 (*d*) or (B10×B10.D_2)F_1 lymphoid cells (*e*), mated to normal B10 females. All tolerant males used for breeding were specifically unresponsive to the tolerizing antigenic determinants in 5-day CML tests (performed in spleen lymphocytes obtained by partial splenectomy under ether anaesthesia) and retained a skin graft of the appropriate F_1 hybrid for at least 100 days (while rejecting third-party grafts in the expected fashion). No data from first-generation progeny born to animals not tolerant by these criteria are shown. Note that the efficiency of inducing stable tolerance in the present strain combinations was lower than in the previous study using (CBA×A)F_1→CBA (ref. 1). Thus, of 9 male and 4 female B10 mice injected at birth with (B10×B10.D_2)F_1, 4 males and 2 females scored tolerant by CML tests and 4 males and 0 females scored tolerant by skin grafting. Of 8 male and 5 female B10 injected at birth with (B10×B10.BR)F_1, 4 males and 1 female were tolerant by CML and 4 males and 0 females were tolerant by skin grafting. Data shown represent cumulative data for progeny of all the tolerant (normal) fathers of a given type (at least 1 litter per father; minimum litter size, 4 offspring). Normal B10 mice tested were age-matched with tolerant progeny and were produced from mating seven normal male B10 mice with normal B10 femals. *f* Shows a limited study (22 mice) of the offspring to B10 mice inoculated neonatally with (B10×B10.D_2)F_1 cells but shown thereafter (by CML and skin graft testing) to be fully responsive to B10.D_2 alloantigens. Offspring from all five males are included in this study (minimum litter size, 3). Cytotoxic data for individual mice were derived from 4-h ^{51}Cr release tests with an effector : target cell ratio of 2 : 1. total releasable c.p.m. and spontaneously released c.p.m. (arithmetic mean ± s.e.m.) for the different targets used were: B10.BR: 9,161 ± 139, 633 ± 11; B10.D_2: 10,132 ± 129, 931 ± 17; ATH: 8,149 ± 101, 576 ± 10. Assays for all targets and all effector populations were performed in triplicate. To test for significance between responses of normal B10 mice and progeny, response groups were compared using 2×2 or 2×3 χ^2 tables. (For 2×2, responses grouped as ≤10%, >10% or ≤20%, >20% specific cytotoxicity; for 2×3, ≤15%, 15–20%, >20% specific cytotoxicity.) χ^2 and probability values are: response to B10.BR: *c* versus *d*, 29.7, $P<0.001$; *c* versus *e*, 2.8, $P>0.2$; response to B10.D_2: *c* versus *d*, 4.4, $P>0.1$; *c* versus *e*, 28.6, $P<0.001$; response to ATH: *c* versus *d*, 0.048, $P>0.8$; *c* versus *e*, 1.2, $P>0.2$.

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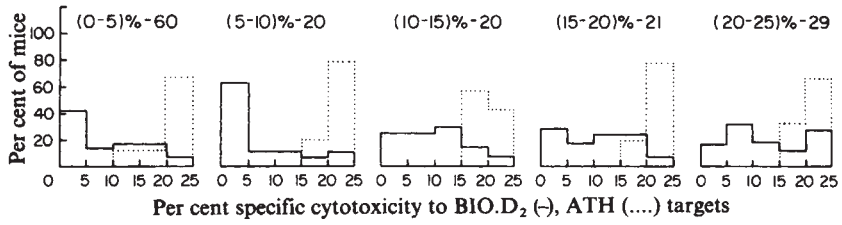

Fig. 2 CML response profile of first-generation progeny born to B10 males tolerized as neonates to both $(B10 \times B10.D_2)F_1$ and $(B10 \times B10.BR)F_1$ lymphoid cells. In this case B10 neonates were inoculated intraperitoneally with a 1 : 1 mixture of 50×10^6 bone marrow cells from the respective F_1 hybrid donors, and these inoculations were repeated every 2 weeks throughout the study. Spleen cells from 8-week-old progeny of matings of B10 animals known to be tolerant to both haplotypes (acceptance of skin grafts from both parents; unresponsiveness in CML cultures of spleen cells obtained by partial splenectomy) were stimulated in culture with irradiated $(B10 \times B10.BR)F_1$, $(B10 \times B10.D_2)F_1$ or ATH spleen cells and cytotoxic responses determined 5 days later. Individuals were then grouped according to their level of cytotoxic response to one tolerizing antigen (B10.BR) and the per cent of mice responding with a given degree of cytotoxicity (0–5%, 5–10% and so on) to the other two antigens recorded. Note (see Fig. 1 legend) that of 19 males and 8 females injected at birth with the bone marrow cell mixture, 6 males and 4 females were tolerant to both $B10.D_2$ and B10.BR in CML assays, and 4 males and 2 females were tolerant to both by skin grafting. As described in Fig. 1 legend, offspring from all 4 males are included in the data shown here (at least 1 litter per male; minimum litter size, 4 offspring). As in Fig. 1, cytotoxic assays were performed in triplicate at an effector: target cell ratio of 2 : 1. Total releasable c.p.m. and spontaneously released c.p.m. (arithmetic mean ± s.e.m.) for the targets shown were: B10.BR: 8,104 ± 106, 609 ± 11; $B10.D_2$: 9,112 ± 113, 613 ± 13; ATH: 7,048 ± 96, 596 ± 14. To test the null hypothesis that the response to $B10.D_2$ is independent of the response to B10.BR, a 3 × 3 table was constructed (response levels being ≤5%, 5–15%, >15% specific cytotoxicity). The $\chi^2 = 8.4$ and $0.05 < P < 0.1$ (null hypothesis cannot be rejected). The group significantly lower than expected (disturbing the rest of the table) was that group responding high to B10.BR (>15%) and low to $B10.D_2$ (≤5%), which contained fewer animals (11) than expected (18.7).

somatically acquired tolerance to two different H–2-encoded haplotypes is assorted at random in the first generation but that a high percentage show evidence for simultaneous transmission of these two specific tolerant traits.

Finally we examined the effect of breeding from the first-generation offspring—derived originally from male B10 mice tolerant to both $B10.D_2$ and B10.BR—on the subsequent transmission of the tolerant traits to second-generation offspring (in all cases only the original grandfathers were ever deliberately exposed to the tolerizing antigenic determinants). Three types of first-generation progeny were used as breeders: those which scored individually tolerant to B10.BR or $B10.D_2$ determinants (Fig. 3*a*, *b*) and those tolerant to both determinants (Fig. 3*c*). These assignments of parental phenotype were made on the basis of CML assays with spleen cells obtained by partial splenectomy.

The data of Fig. 3*a*, *b*, although limited to studies of only 27 and 15 individuals respectively, are clearly quite different from those of Fig. 2, and are, in fact, similar to those obtained by re-plotting the data of Fig. 1*d*, *e* in this format. Thus, first-generation progeny parents phenotypically tolerant to B10.BR give rise to second-generation offspring which respond to $B10.D_2$ antigens, yet which have retained their unresponsive phenotype for B10.BR antigens (Fig. 3*a*). Similar effects are also seen in the second-generation progeny derived from first-generation parents tolerant to $B10.D_2$ antigens (Fig. 3*b*). No unexpected reacquisition of tolerance for the other MHC haplotype was observed in any of the 42 mice tested (see below). In contrast to these patterns, first-generation progeny breeders scoring tolerant to both B10.BR and $B10.D_2$ antigens give rise to second-generation progeny (Fig. 3*c*) with the same 'spray' of cytotoxic response patterns as seen with the initial first-generation progeny (compare with Fig. 2). Some animals were tolerant either to B10.BR or $B10.D_2$ antigens, some were still tolerant to both antigens, and some had lost tolerance to both antigens.

In summary, investigation of the inheritance of neonatally acquired tolerance to major histocompatibility complex (MHC) gene products in congeneic resistant mice (using a B10 background) has shown that two separately determined tolerance phenotypes can be transmitted and assorted independently in first-generation progeny derived from matings of males tolerized to two H–2 haplotypes with normal females; and moreover, that a significant proportion of these first-generation progeny are also unresponsive towards determinants encoded by genes associated with both haplotypes. Transmission of these unresponsive traits is relatively stable as indicated by the fact that no spontaneous reversion (to the 'double' tolerance state) was observed in second-generation progeny which failed to display tolerance to one of the two H–2 haplotypes, and a significant percentage (16%, $P < 0.05$; see left-hand panels of Fig. 3*c*) of second-generation mice born to parents tolerant to both haplotypes still fail to respond to both, although independent assortment of the two responses can also be observed. These results suggest that genetic transmission of acquired tolerance to H–2 antigens may assort in a mendelian manner.

We noted what may be a more apparent than real dichotomy in the independent transmission of tolerance to $H\text{–}2^k$, $H\text{–}2^d$ haplotypes (from mice tolerant of B10.A), yet the simultaneous transmission of unresponsiveness to $H\text{–}2K^k$, $H\text{–}2D^k$ from mice tolerant of B10.BR. As we pointed out, we have not rigorously tested the response of mice to either K^k or D^k determinants (although independent transmission of tolerance to K^k, D^d does occur from B10 mice tolerant to B10.A MHC determinants, unpublished results). We reported previously a phenomenon of 'skipping' of expression of the tolerance phenotype in sequential generations of CBA mice made tolerant to $(CBA \times A)F_1$ antigens[1]. Thus second-generation tolerant progeny are derived from apparently normal responder first-generation mice. In contrast, we recorded (Fig. 3) here a failure to detect 'reversion' or skipping) in sequential generations of mice. We cannot yet explain these differences, but it is not unreasonable to anticipate that they are due to inherent differences in the two experimental situations: for instance, to the ease with which tolerance can be produced in CBA (to A/J antigen) relative to the difficulty reported here in tolerizing B10 mice.

Finally, note the apparent difference in the behaviour (as assessed by CML response) of progeny mice derived from mating A-strain females with either A males neonatally tolerized to $(A \times B)F_1$ cells or normal $(A \times B)F_1$ males. In the latter case $(A \times A)$, offspring are not unresponsive to B antigens, despite the fact that tolerance to both A and B in F_1 mice presumably takes place after fusion of gametes. Nevertheless, the data presented here and elsewhere show that in the former situation the progeny acquire the tolerance phenotype of their parents. We cannot yet explain convincingly this anomaly; however, there are apparently many mechanisms whereby somatically acquired tolerance may be induced and maintained[7], and so far there is little evidence to indicate which, if any, of these mechanisms represents the natural mode of self/non-self discrimination.

The high frequency of parallel transmission of independently assorted, somatically acquired characters severely challenges Weismann's doctrine, and seems to us to demand an explanation not offered by conventional neo-darwinian genetics. It has been

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argued[2] that the type of phenomenon documented here and elsewhere[1,2,8,9], may be explained best by invoking the clonal selection and somatic mutation theory of immunology[10–13] and soma germ-line transmission of somatic genetic information (RNA) through endogenous RNA viral vectors and reverse transcriptase[14,15]. We might then imagine that genetic transfer of somatically selected information associated with the regulation of responses, for example, to A/J antigens in CBA mice[1] (anti-idiotype ?, antigen ?) is crucial for the anomalous inheritance patterns observed.

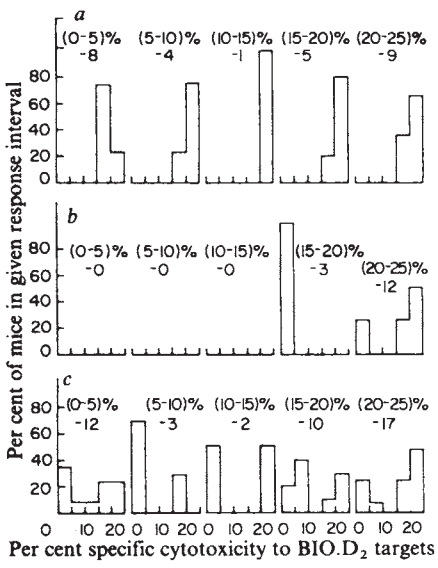

Fig. 3 Analysis of transmission of response patterns from first-generation progeny (Fig. 2) to second-generation progeny. Three types of first-generation progeny were used as parents to produce this data. In *a*, animals were selected (by CML tests performed after partial splenectomy) which were tolerant only to B10.BR stimulator cells—these first-generation progeny gave normal responses after stimulation with $(B10 \times B10.D_2)F_1$ cells. *b* Shows second-generation progeny derived from first-generation animals found to be tolerant only to $B10.D_2$ antigens. The data of *c* were derived from second-generation progeny born to animals found to have inherited the tolerance phenotype of the original male parents unresponsive to both $B10.D_2$ and B10.BR antigens). In all cases breeding was performed by mating either phenotypically tolerant males/females to normal females/males (outcross) or by incrossing of first-generation progeny with identical CML response phenotypes (earlier studies have suggested little difference in the gross inheritance of tolerance from such matings)[1]. Thus, for *a* the data are pooled from 5 litters from incross matings and 1 litter (5 progeny) of an outcross first-generation female; for *b* the data are pooled from 3 litters of 3 incross matings, while for *c* the data represent cumulative studies on 10 litters ($\geq$1 per cage; minimum size, 4 mice) of 4 incross matings and 3 outcrosses (using first-generation tolerant female). The data of these second-generation progeny (total individual numbers tested for *a*, *b*, *c*, were 27, 15, 44 respectively) were grouped into response intervals for one antigen and the frequency of response in a given cytotoxic interval for the other antigen plotted (see Fig. 2). As in Figs 1, 2, cytotoxic responses after stimulation with ATH cells were also determined, but the pattern of response was not qualitatively or quantitatively different from that shown in Figs 1, 2 (all animals responded, irrespective of response status to $B10.D_2$ or B10.BR, data not shown). All data were obtained from the assays performed in triplicate at an effector:target cell ratio of 2:1. Total releasable c.p.m. and spontaneously released c.p.m. (arithmetic mean ± s.e.m.) for the different targets used were: B10.BR: 9,100 ± 113, 689 ± 16; $B10.D_2$: 7,068 ± 94, 495 ± 12; ATH: 10,351 ± 129, 689 ± 21. To test the null hypothesis that the response to $B10.D_2$ is independent of the response to B10.BR (*c*), a 2×2 table was constructed ($\leq$15%, >15%) and an exact χ^2 test performed giving $P > 0.2$ (null hypothesis cannot be rejected).

We thank Susannah MacRae, Barbara Khomasurya, Susan Oliphant and Sue Bejasa for assistance. For critical comments on the manuscript we thank Drs Elizabeth Simpson, Bob Blanden, Arno Mullbacher and Jeff Pollard; and Walter Fierz and Steve Molnar for advice on statistics. This work was supported by the MRC of Canada (grant MA 5440) and the NCI of Canada. E.J.S. thanks the KIB Foundation (UK) for support.

Received 11 June; accepted 3 December 1980.

1. Gorczynski, R. M. & Steele, E. J. *Proc. natn. Acad. Sci. U.S.A.* **77**, 2871–2875 (1980).
2. Steele, E. J. *Somatic Selection and Adaptive Evolution* (Williams & Wallace, Toronto; Croom Helm, London, 1979).
3. Billingham, R. E., Brent, L. & Medawar, P. B. *Phil. Trans. R. Soc.* B**239**, 357 (1956).
4. Gorczynski, R. M., MacRae, S. & Till, J. E. *Scand. J. Immun.* **7**, 453–465 (1978).
5. Gorczynski, R. M. & MacRae, S. *J. Immun.* **122**, 737–746 (1979).
6. Gorczynski, R. M. *Immunology* **33**, 697–712 (1977).
7. Gorczynski, R. M., Khomasurya, B., Short, L. & MacRae, S. in *Regulation by T Cells* 18 (University of British Columbia Press, 1979).
8. Guttman, R. D. & Aust, J. B. *Nature* **197**, 1220–1221 (1963).
9. Kanazawa, R. D. & Imai, A. *Jap. J. exp. Med.* **44**, 227–234 (1974).
10. Burnet, F. M. *The Clonal Selection Theory of Acquired Immunity* (Cambridge University Press, London, 1959).
11. Cohn, M. *Progress in Immunology II* Vol. 1 (eds Brent, I. L. & Holborow, J.) 261–284 (North Holland, Amsterdam, 1974).
12. Jerne, N. K. *Eur. J. Immun.* **1**, 1–9 (1971).
13. Cunningham, A. J. *Contemp. Topics molec. Immun.* **3**, 1–26 (1974).
14. Temin, H. M. *J. natn. Cancer Inst.* **46**, 3–8 (1971).
15. Temin, H. M. *Science* **192**, 1075–1080 (1976).

B220: a B cell-specific member of the T200 glycoprotein family

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T200, a major cell-surface glycoprotein on lymphoid cells, exists in several forms with different electrophoretic mobilities. These forms have been correlated with different classes of lymphoid cell. The smaller forms, with molecular weights (MWs) of ≃170,000 and 180,000, are found predominantly on T cells while the 220,000 MW form is associated with B cells[1,2]. The polypeptide portions of each molecule may be identical or closely related as all three forms share the same allelic variations[3–5], and all reported heterologous antisera and monoclonal antibodies to T200 precipitate all three forms[2,6]. We report here a monoclonal antibody specific for the 220,000 MW form of T200 and show that it is expressed only on B cells and a subset of bone marrow cells which includes B cell precursors. We suggest that this form of the molecule be designated provisionally B220.

The hybridoma RA3-3A1 was produced by the fusion of the mouse myeloma S194/5.XX0.BU-1 with spleen cells from a rat immunized with the mouse cell line RAW 112, a pre-B cell tumour derived by infection of a mouse with the Abelson murine leukaemia virus[7]. The monoclonal antibody-secreting hybridoma RA3-3A1 was selected from the other fusion products because it produced an antibody that recognized RAW 112 cells and a subset of bone marrow cells, but not thymocytes. About 30% of bone marrow cells, 50% of spleen cells, 40% of mesenteric lymph node cells and less than 2% of thymocytes were positive when stained with the optimum titre of RA3-3A1 followed by fluorescein-conjugated rabbit anti-rat immunoglobulin. Flow cytometry analysis was carried out using the fluorescence-activated cell sorter (FACS III, Becton Dickinson, California). RA3-3A1 is an IgM and binds directly to the bacterium *Staphylococcus aureus*.

Immunoprecipitation of surface-iodinated spleen and RAW 112 cell lysates followed by SDS-polyacrylamide gel electrophoresis (PAGE) showed that RA3-3A1 precipitated a molecule with an apparent MW of 220,000 (Fig. 1). Precipitation of the same lysates with a monoclonal anti-T200 antibody gave the expected three bands from the spleen, the

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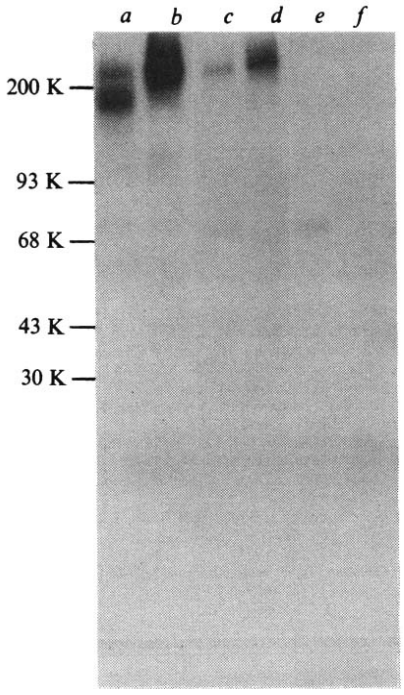

Fig. 1 Immunoprecipitation with RA3-3A1 and anti-T200. BALB/c spleen cells and RAW 112 cells were surface-iodinated by the lactoperoxidase method[8]. The cells were lysed with 1% NP40 in 0.01 M sodium phosphate buffer, *p*H 7.5, and the lysates clarified at 100,000*g* for 1 h. Aliquots of lysate from 2×10^6 cells were incubated with monoclonal antibodies and then precipitated with *S. aureus*[9] (for RA3-3A1 and control IgM) or *S. aureus* saturated with purified rabbit anti-rat Ig antibodies (for anti-T200). The rat anti-T200, prepared from the hybridoma 55-10.2 (ref. 10), was given by Dr J. A. Ledbetter. Control precipitates were prepared from rat IgM monoclonal antibody with no apparent reactivity to mouse cells. Immune precipitates were washed three times in 0.05 M Tris, *p*H 8.3, 0.45 M NaCl and 0.5% NP40, boiled in sample buffer with 1% β-mercaptoethanol and electrophoresed on 5–20% gradient SDS-polyacrylamide gels. The gels were run at 125 V for 1 h after the bromophenol blue marker dye ran off the end of the gel. The gels were then dried, fixed and autoradiographed on Kodak X-R film. Lanes *a*, *c* and are spleen cells; lanes *b*, *d* and *f* are RAW 112 cells; *a* and *b* *b* are precipitated with anti-T200, *c* and *d* with RA3-3A1, and *e* and *f* with control rat IgM. Molecular weights are expressed in thousands (K).

Table 1 Identity of RA3-3A1$^+$ and RA3-2C2$^+$ populations in bone marrow and spleen

Cell fraction	%Cells stained with: RA3-3A1	RA3-2C2	Anti-κ
Whole bone marrow	32	28	10
RA3-3A1$^-$ bone marrow	3	3	1
RA3-2C2$^-$ bone marrow	2	2	1
sIg$^-$ bone marrow	19	19	3
Whole spleen	37	40	45
RA3-3A1$^-$ spleen	1	3	2
RA3-2C2$^-$ spleen	4	3	3
sIg$^-$ spleen	4	5	2

Spleen and bone marrow cells from BALB/c mice were stained with the appropriate antibody followed by a second stage with fluorescein-conjugated rabbit anti-rat Ig (for RA3-3A1 and RA3-2C2) or goat anti-rabbit Ig for the rabbit anti-mouse-κ. The reagents and details of staining and sorting procedures are described elsewhere[11]. The unstained fraction was isolated on the FACS III and samples restained as indicated. Sorted fractions contained 1–3% residual positive cells. The figures represent the per cent positive with the specific antibody minus the per cent positive in a control stain. The control for the rat monoclonals was an unreactive rat IgM monoclonal; for the anti-κ, it was specifically eluted rabbit anti-keyhole-limpet haemocyanin.

largest of which co-migrated with the molecule recognized by RA3-3A1. For RAW 112 cells the anti-T200 precipitated only one wide band centred around MW 220,000, which co-migrated with the band precipitated by RA3-3A1.

To test whether or not the 220,000 MW protein recognized by RA3-3A1 was the same as the large, B cell-associated form of T200, a surface-iodinated spleen cell lysate was precleared with anti-T200 or RA3-3A1 before immunoprecipitation and SDS-PAGE analysis with the two antibodies. Preclearing with anti-T200 removed all the bands precipitable by anti-T200 and also removed all the RA3-3A1-precipitable material (Fig. 2). However, preclearing with RA3-3A1 removed almost all the RA3-3A1-precipitable material, but seemed to remove only part of the 220,000 MW band. This is consistent with the observation (in Fig. 1) that RA3-3A1 does not precipitate as much 220,000 MW material as does anti-T200. This could mean that the 220,000 MW band precipitated by anti-T200 represents two or more antigenically distinct molecules, not all of which are recognized by the RA3-3A1 antibody. Alternatively, the determinant recognized by RA3-3A1 may be more labile than that recognized by the anti-T200, or the RA3-3A1 antibody may have a lower affinity for the molecule than the anti-T200 antibody. The precipitation of comparable amounts of μ chains from untreated and precleared lysates demonstrated that an unrelated surface glycoprotein was not removed by preclearing.

RA3-3A1 was shown to be specific for B cells and B cell precursors by comparing its staining distribution in the spleen and bone marrow with that of an anti-κ antiserum and another monoclonal antibody, RA3-2C2, which has been shown to be specific for B cells in peripheral lymphoid tissues and for B cells

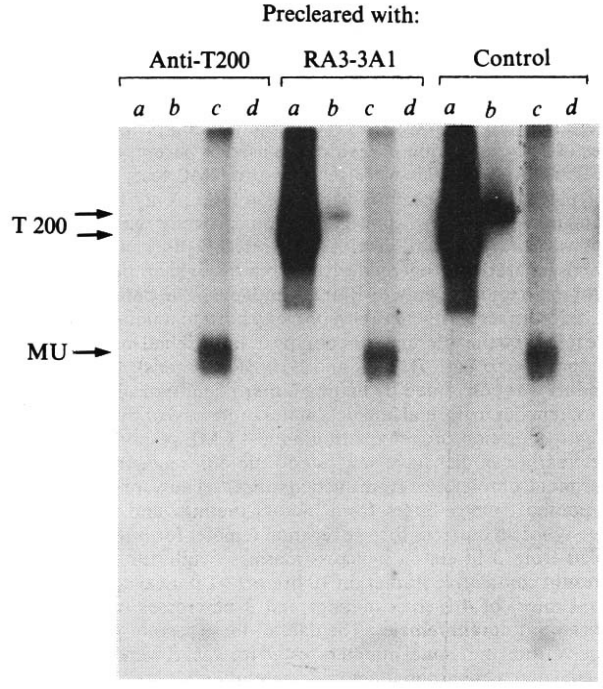

Fig. 2 Preclearing of surface-iodinated spleen lysate with RA3-3A1 and anti-T200. Surface-iodination, antibody precipitation and gel electrophoresis were performed as described in Fig. 1 legend. Preclearing of lysate aliquots was carried out before precipitation by adding a large excess of antibody to the lysates followed by two cycles of precipitation with *S. aureus* (for RA3-3A1 and control IgM) or *S. aureus* saturated with rabbit anti-rat Ig (for anti-T200). Precleared lysates representing 4×10^6 spleen cells were split into four aliquots and immunoprecipitated with: *a*, anti-T200; *b*, RA3-3A1; *c*, monoclonal rat anti-mouse μ chain; and *d*, control rat IgM. The T200 bands were deliberately overexposed to demonstrate the extent of the preclearing. Rat anti-mouse μ chain, produced from the hybridoma 151–119, was given by Dr J. Haaijman.

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and sIg$^-$ lymphoid cells in the bone marrow[11]. Removal of sIg$^+$ or RA3-2C2$^+$ cells from the spleen by cell sorting removes all RA3-3A1$^+$ cells and, conversely, removal of the RA3-3A1$^+$ cells removes all cells that are sIg$^+$ and RA3-2C2$^+$ (Table 1). Thus, the molecule recognized by RA3-3A1 is expressed on all splenic B cells. A similar experiment with bone marrow cells again shows that RA3-3A1 and RA3-2C2 recognize the same population of cells, which includes B cells and ~20% of the sIg$^+$ bone marrow cells (Table I). These RA3-2C2$^+$, sIg$^-$ bone marrow cells include those which differentiate into sIg$^+$ cells in two days *in vitro*[11]. RA3-3A1 is therefore expressed on a fraction of sIg$^-$ bone marrow cells which consists of small lymphocytes and medium and large blast cells, and which includes at least some B cell precursors. Despite the similar staining properties of the two monoclonal antibodies, they seem to recognize different molecules. RA3-2C2 precipitates no molecules in the 200,000 MW range from surface-iodinated spleen cells, but it does precipitate a 50,000 MW protein from lysates of biosynthetically labelled lymphoid cells or tumours[12]. This p50 protein is not precipitated by RA3-3A1.

Because the form of T200 recognized by RA3-3A1 is expressed predominantly if not entirely by cells in the B cell lineage, we suggest that this molecule provisionally be called B220, and that the name T200 be reserved for the molecules found on thymus-derived lymphocytes.

We thank Dr William Raschke for the RAW 112 cell line, Drs J. Ledbetter and J. Haaijman for antibodies, and Dr Ian Trowbridge for helpful discussions. This work was supported by NCI fellowships CA 09302 and CA 06429 to R.L.C. and grants CP 91011 from the NCI, AI 09072 from the NIH and IM 56 from the American Cancer Society.

Received 9 October; accepted 2 December 1980.

1. Trowbridge, I. S., Ralph, P. & Bevan, M. J. *Proc. natn. Acad. Sci. U.S.A.* **72**, 157–161 (1975).
2. Trowbridge, I. S. & Mazauskas, C. *Eur. J. Immun.* **6**, 557–562 (1976).
3. Michaelson, J., Scheid, M. & Boyse, E. A. *Immunogenetics* **9**, 193–197 (1979).
4. Omary, M. B., Trowbridge, I. S. & Scheid, M. P. *J. exp. Med.* **151**, 1311–1316 (1980).
5. Siadak, A. W. & Nowinski, R. C. *J. Immun.* **125**, 1400–1401 (1980).
6. Trowbridge, I. S. *J. exp. Med.* **148**, 313–323 (1978).
7. Ralph, P., Nakoinz, I. & Raschke, W. C. *Biochem. biophys. Res. Commun.* **61**, 1268–1275 (1974).
8. Fox, R. I. & Weissman, I. L. *J. Immun.* **122**, 1697–1704 (1979).
9. Kessler, S. W. *J. Immun.* **115**, 1617–1624 (1975).
10. Ledbetter, J. A. & Herzenberg, L. A. *Immun. Rev.* **47**, 63–90 (1979).
11. Coffman, R. L. & Weissman, I. L. *J. exp. Med.* (in the press).
12. Rotter, V., Witte, O. N., Coffman, R. & Baltimore, D. *J. Virol.* **36**, 547–555 (1980).

Induction of cell–cell channel formation by mRNA

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Intercellular junctional communication is very common in normal organized tissue[1]. It provides a pathway for transmission of electrical signals, especially in heart muscle[2,3], and may be important in differentiation and growth control[1,4]. The hydrophilic channels which enable cell–cell communication have been well characterized by biophysical methods[5,6], and there is now good evidence that they are contained in the nexus (gap junctions)[3,7–9]. Little, however, is known about the molecular mechanism of biosynthesis of junctional channels. Knowledge in this area has been obtained almost exclusively from experiments with reaggregated cells[10–15], a system complicated by the fact that *de novo* synthesis of channel proteins is obscured by reassembly of pre-existing subunits or utilization of precursors. To avoid these problems, we have now isolated mRNA from cells that are in the process of making new intercellular nexus with high efficiency, incorporated it via liposomes into communication-defective cells and have shown that the recipient cells established junctional communication.

We wanted a system where formation of junctions is controlled by an external stimulus and so chose uterine smooth muscle cells, because their response to stimulus is more pronounced than in the few other cell types where induction has been observed[16–20]. Normally, the cells of the myometrium do not exhibit synchronous contractile activity and have very few gap junctions. By contrast, during parturition when well synchronized activity of the cells of the myometrium is required for expulsion of the fetus, numerous large gap junctions are observed[16,17]. Although we have already reported that nexus formation in the myometrium can be induced in virginal females by injection of oestrogen[16], it was not clear whether this was due to *de novo* synthesis of junctional elements or to reassembly of pre-existing subunits. On the assumption that we were observing *de novo* synthesis of gap junction proteins[21–23] there should be present in these cells an increased number of mRNA molecules coding for these proteins shortly after stimulation with oestrogen.

To investigate this prediction we first obtained either a polysomal fraction or free mRNA from oestrogen-treated uterine tissue of rats. The polysomal fraction, or free mRNA, was then encapsulated into liposomes by the reverse-phase evaporation technique[24]. The phospholipid composition (phosphatidylserine/phosphatidylcholine) of the liposomes was chosen to ensure liposome–cell membrane fusion. Control experiments with liposomes containing fluorescent dyes, and electron microscopy, have shown that fusion was indeed a predominant form of liposome–cell interaction (R.A and G.D., unpublished results) (Fig. 1).

To test whether the injected mRNA coded for gap junction protein we measured its ability to induce intercellular communication between cells of the communication-defective Cl-1D mouse cell line[25]. In all experiments confluent cultures of

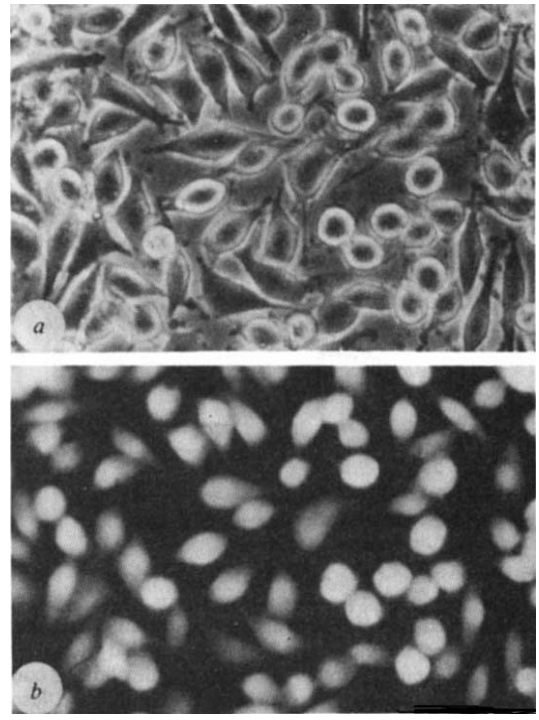

Fig. 1 Appearance of Cl-1D cells in bright field (*a*) and dark field (*b*) after incubation for 10 min with liposomes containing the fluorescent tracer substance carboxyfluorescein. The liposomes were prepared as described for the encapsulation of polysomes except that 100 mM carboxyfluorescein replaced the polysomes. All cells are highly fluorescent and even small cell processes contain sufficient tracer substance to outline them clearly.

0028–0836/81/080683—03$01.00

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Cl-1D cells were first washed with serum-free minimal essential medium (MEM). The liposome preparation was diluted 1:3 (v/v) with serum-free MEM, and 1 ml of this suspension was added to a 9.6-cm^2 dish of Cl-1D cells. The incubation period was 1 h at 37 °C. The liposomes were then removed, and the cells washed repeatedly and incubated in normal culture medium. Thereafter random cell pairs were tested for electrical coupling (Fig. 2). Most cell pairs when tested 6 h after the removal of the liposomes were found to be electrically coupled (Table 1). The observed coupling ratios varied, but were consistently <1.0. Control cultures of Cl-1D cells were incubated in identical conditions with liposomes containing buffer only; no coupling was observed. Similarly, if the polysome preparation was treated with ribonuclease before liposomal encapsulation, no coupling developed. When cycloheximide was added to the cells immediately after the 1-h incubation period with mRNA-containing liposomes, the expression of the mRNA was drastically reduced; only 1 out of 13 cell pairs showed any coupling.

Although most of our experiments were done with polysome preparations we have obtained similar results with free mRNA isolated from oestrogen-induced uterine cells (Table 1). In contrast to polysomes, however, free mRNA had to be encapsulated at higher concentrations than polysomes to be as effective in inducing cell–cell coupling. This was not unexpected because the free mRNA was extracted from total cellular RNA whereas polysomes were obtained from a microsomal fraction known to be enriched for mRNA coding for membrane proteins. Thus, the free mRNA may have a lower specific activity than polysomes to induce cell–cell coupling.

Studies of the time course of development of cell–cell coupling showed that coupling appeared within 2–4 h after removal of the liposomes (Table 2). At 4 h, about 40% of the cells were coupled, and this percentage increased still further until at 8 h practically all cells were coupled. The increase in coupling incidence was accompanied by larger coupling ratios. Later, the frequency of coupling declined and disappeared completely at about 30 h.

Freeze-fracture electron microscopy[29] of mRNA-treated Cl-1D cells revealed the presence of nexus-like structures at the time when cell–cell coupling was present (Fig. 3). The observed structures consisted of aggregates of membrane particles resembling nexus which were associated with short ridges appearing like tight junctions. Similar structural arrangements have been found in myometrial tissue during early stages of nexus development after oestrogen treatment (G.D., unpublished results).

To determine whether the observed development of electrical coupling between Cl-1D cells occurs through newly synthesized channel proteins, the following potential artefacts must be ruled out. First, coupling could have developed because of fusion or partial fusion between neighbouring cells. Second, our polysome preparation may have been contaminated by channel

Table 1 Incidence of electrical coupling in Cl-1D cells in control conditions and after loading with mRNA

Conditions of liposome treatment	Coupling incidence
Control: liposomes containing buffer only	0/29 (3)
Liposomes containing polysomes	77/114 (14)
Polysomes treated with ribonuclease before encapsulation	0/18 (2)
Cycloheximide (5 $\mu g\ ml^{-1}$) added after polysome loading	1/13 (2)
Free mRNA-containing liposomes:	
5 $\mu g\ ml^{-1}$ mRNA	4/20 (2)
25 $\mu g\ ml^{-1}$ mRNA	17/22 (2)

The number of cell pairs exhibiting electrical coupling (first number) and the total number of cell pairs impaled with microelectrodes and exhibiting stable membrane potentials (second number) show the coupling incidence. The number of experiments is given in parentheses. Coupling incidence was determined 6 h after treatment of the cells with liposomes. For obtaining the mRNA preparations, mature female virgin rats (Sprague–Dawley) were injected intramuscularly with oestrogen (Delestrogen, Squibb). Each animal received 10 mg of oestrogen per day for 3 days. On the fourth day the rats were killed and the uteri removed. (In some experiments the endometrium was removed by microdissection.) The uterine tissue from five rats was homogenized with an Ultra Turrax (Tekmar) homogenizer in 15–20 ml of buffer A (25 mM sucrose, 1.5 mM $MgCl_2$, 50 mM NaCl, 25 mM HEPES, *p*H 7.2). After centrifugation for 3 min at 5,000*g*, the supernatant was further centrifuged at 100,000*g* for 60 min. The pellet obtained in the high-speed centrifugation (microsomal fraction) was resuspended in 2 ml of buffer B (25 mM NaCl, 5 mM $MgCl_2$, 25 mM HEPES, *p*H 7.2) containing 0.5% (v/v) NP40 detergent. After incubation at 0 °C for 60 min and removal of insoluble fragments by centrifugation (10,000*g*, 30 min), the solution was layered on a discontinuous sucrose gradient containing 1 ml 2.5 M sucrose (in buffer B) and 2 ml 1.0 M sucrose (in buffer B), and centrifuged in a Spinco SW 50.1 rotor at 43,000 r.p.m. for 3 h. A polysomal fraction was recovered from the interface between the two sucrose layers. The polysomes were dialysed overnight against buffer C (1 mM EGTA, 15 mM KCl, 10 mM NaCl, 5 mM HEPES, *p*H 7.2), concentrated by freeze-drying to about 1 ml and dialysed again against buffer C containing 150 mM KCl. The volume of the final polysome preparation was adjusted to a concentration of $A_{260}=2.0$–3.0 (ratio $A_{260}/A_{280}=1.5$–1.6). Free mRNA was isolated by homogenization of uterine tissue in 4 M guanidine thiocyanate according to the procedure by Chirgwin *et al.*[27]. All equipment and containers used in these experiments were treated with dietyl pyrocarbonate (0.1%) for inactivation of RNase. The mRNA was further purified by adsorption to oligo(dT) cellulose[28]. For encapsulation of the mRNA preparations into liposomes, the reverse-phase evaporation technique[24] was used with the following specifications: 20 mg of phosphatidylserine and phosphotidylcholine (ratio 3:2) per 0.5 ml of aqueous solution of RNA. The liposome preparation was dialysed against Ca, Mg-free Hank's balanced salt solution (Gibco).

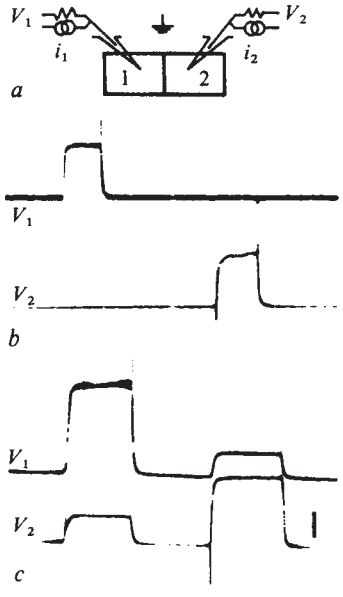

Fig. 2 Measurements of electrical cell–cell coupling. *a*, Electrode arrangement. Two neighbouring cells were impaled with a microelectrode each, which through a bridge circuit allowed current injection and recording of membrane potential. Oscilloscope traces: *b*, membrane potential in Cl-1D cells in control conditions; cells were incubated with liposome-containing buffer solution only. Current injection (10^{-8} A) into cell 1 results in hyperpolarization of this cell (V_1), which does not result in a voltage change in cell 2 (V_2). After a delay of about 150 ms, current is injected into cell 2, which results in change of membrane potential in this cell but not in cell1. *c*, Membrane potential Cl-1D cells incubated with liposomes containing the mRNA preparation. Current injection in either one of the cells results in potential changes in both cells indicating electrical coupling. Calibration 50 mV.

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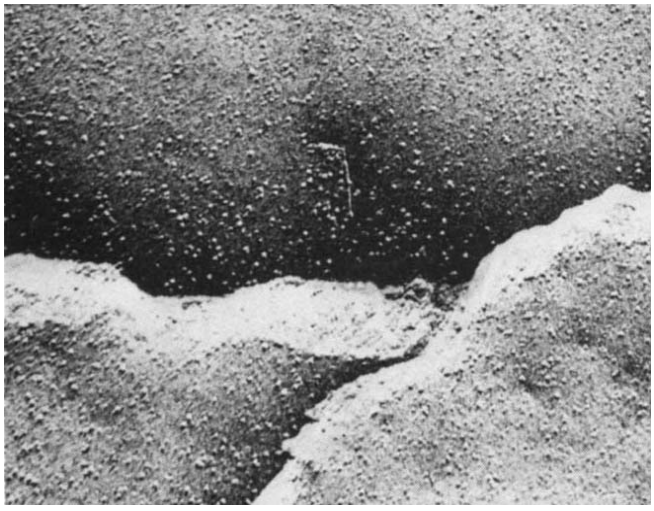

Fig. 3 Freeze-fracture electron micrograph of Cl-1D cells 6 h after incubation with liposomes containing the mRNA preparation. Within an area of membrane apposition a small aggregate of membrane particles can be seen associated with short ridges. ×60,000.

proteins of the uterine cells. Third, the liposome treatment by itself may have nonspecifically induced the development of cell–cell coupling in the recipient cell. Cell–cell fusion can be ruled out, as in all our experiments the observed coupling ratios were considerably below 1.0, whereas fusion should result in coupling ratios of 1.0 or close to 1.0, as has been shown with fusing myoblasts[26]. Furthermore, the control experiments with ribonuclease or cycloheximide indicate that mRNA is required and that protein synthesis is involved in the establishment of coupling. This argument is strengthened by the results obtained with free mRNA. Liposomes by themselves did not produce coupling. The time course of the development of coupling also seems to argue against an artefact. Polysomes from uterine cells that had not been treated with oestrogen did not produce any coupling. Finally, freeze-fracture electron micrographs show membrane junctions. Although the observed structures are not typical for nexus they resemble the structures that can be seen in the uterus shortly after oestrogen treatment. It is possible that the mRNA concentration is too low to induce fully developed nexus. This possibility is supported by the observed coupling ratios which are much lower than those observed in normal, coupled cells. Alternatively, junction formation may be a complex process requiring the participation of several factors some of which may not be under oestrogen control and, consequently, their mRNA may not be present at a high enough concentration in the polysome fraction.

It seems likely, therefore, that functional channels are present in the mRNA-treated Cl-1D cells but are not easily identified by electron microscopy because of the absence of typical multi-unit structures. Assuming that we are observing formation of regular cell–cell channels, we may conclude that their functional lifetime is of the order of 24 h. This seems to be a reasonable estimate because nexus have been observed for a similar period in myometrial cells at term[16]. Of course, the functional lifetime of intercellular channels in organized tissue may be quite different from that observed in cultured cells which are rapidly proliferating and, therefore, have a higher overall turnover rate. In conclusion, our results suggest that the induction of cell–cell coupling in rat uterus smooth muscle by oestrogen occurs by *de novo* synthesis of cell–cell channel proteins. Although this is the most straightforward interpretation of our results, we realize that alternative possibilities cannot be ruled out. For example, the introduced mRNA might code for a regulatory or morphopoietic protein involved in cell–cell communication. We believe that the system described here provides a promising model for obtaining information on the biosynthesis of junctional proteins not readily obtainable by conventional methods.

Table 2 Time course of development of cell–cell coupling after loading with polysomes

Hours after liposome treatment	Coupling incidence
2	0/12
4	4/10
6	9/10
8	10/10
25	3/10
30	0/10

Data obtained from one experiment. Two additional experiments gave similar results.

We thank Dr W. R. Loewenstein for continuous encouragement, and A. Diaz-Calejo and L. Hevia for technical assistance. This research was supported by PHS grants CA 14464, CA 14395 and GM 22510.

Received 21 August; accepted 11 December 1980.

1. Loewenstein, W. R. *Biochim. biophys. Acta* **560**, 1–65 (1979).
2. Weidmann, S. *J. Physiol., Lond.* **118**, 348–360 (1952).
3. Barr, L., Dewey, M. M. & Berger, W. *J. gen. Physiol.* **48**, 797–823 (1965).
4. Loewenstein, W. R. *Devl Biol.* **19**, Suppl. 2, 151–183 (1968).
5. Loewenstein, W. R. *Cold Spring Harb. Symp. quant. Biol.* **40**, 49–63 (1975).
6. Flagg-Newton, J., Simpson, I. & Loewenstein, W. R. *Science* **205**, 404–407 (1979).
7. McNutt, N. S. & Weinstein, R. S. *Prog. Biophys. molec. Biol.* **26**, 45–101 (1973).
8. Caspar, D. L. D., Goodenough, D. A., Malkowsky, L. & Phillips, W. C. *J. Cell Biol.* **74**, 605–628 (1977).
9. Unwin, P. N. T. & Zampighi, G. *Nature* **283**, 545–549 (1980).
10. Loewenstein, W. R. *Devl Biol.* **15**, 503–520 (1967).
11. Ito, S., Sato, E. & Loewenstein, W. R. *J. Membrane Biol.* **19**, 305–337 (1974).
12. DeHaan, R. L. & Hirakow, R. *Expl Cell Res.* **70**, 214–220 (1972).
13. Hammer, M., Epstein, M. & Shridan, J. *J. Cell Biol.* **59**, 130a (1973).
14. Epstein, M. L., Sheridan, J. D. & Johnson, R. G. *Expl Cell Res.* **104**, 25–30 (1977).
15. Griepp, E. B. & Bernfield, M. R. *Circulation* **52**, Suppl. 2, 106 (1975).
16. Dahl, G. & Berger, W. *Cell Biol. int. Rep.* **2**, 381–387 (1978).
17. Garfield, R. E., Sims, S. M., Kannan, M. S. & Daniel, E. E. *Am. J. Physiol.* **235**, C168–C179 (1978).
18. Merk, F. B., Botticelli, C. R. & Albright, J. T. *Endocrinology* **90**, 992–1007 (1972).
19. Decker, R. S. *J. Cell Biol.* **69**, 669–685 (1976).
20. Decker, R. S., Donta, S. T., Larsen, W. J. & Murray, S. A. *J. supramolec. Struct.* **9**, 497–507 (1978).
21. Henderson, D., Eibl, H. & Weber, K. *J. molec. Biol.* **132**, 193–218 (1979).
22. Hertzberg, E. L. & Gilula, N. B. *J. biol. Chem.* **254**, 2138–2147 (1979).
23. Finbow, M., Yancey, S. B., Johnson, R. & Revel, J. P. *Proc. natn. Acad. Sci. U.S.A.* **77**, 970–974 (1980).
24. Szoka, F. & Papahadjopoulos, D. *Proc. natn. Acad. Sci. U.S.A.* **75**, 4194–4198 (1978).
25. Azarnia, R., Larsen, W. J. & Loewenstein, W. R. *Proc natn. Acad. Sci. U.S.A.* **71**, 880–884 (1974).
26. Rash, J. E. & Fambrough, D. *Devl Biol.* **30**, 168–186 (1973).
27. Chirgwin, J. M., Przybyla, A. E., MacDonald, R. J. & Rutter, W. J. *Biochemistry* **18**, 5294–5299 (1979).
28. Kryxtosek, A., Cawthon, M. L. & Kabat, D. *J. biol. Chem.* **250**, 6077–6084 (1975).
29. Pauli, B. K., Weinstein, R. S., Soble, L. W. & Alroy, J. *J. Cell Biol.* **72**, 763–769 (1977).

Block of Na current and intramembrane charge movement in myelinated nerve fibres poisoned with a vegetable toxin

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In nerve membrane, the non-linear capacity current (displacement current) is assumed to reflect the movement of intrinsic membrane charges which control the opening of specific pathways for sodium ions (Na channels). However, various discrepancies have been reported between the effects of pharmacological agents on sodium and displacement currents (for a review see ref. 1). It is generally supposed that the opening and closing of Na channels constitutes one step of a multi-step system in which each configuration change may or not give rise to a measurable charge movement[2]. New drugs affecting either

0028–0836/81/080685—04$01.00

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sodium or displacement currents may elucidate the relationship between charge movement and the control of sodium conductance. We therefore now report a comparison of the effects of a vegetable toxin (oenanthotoxin or OETX)[3] on both sodium current (I_{Na}) and intra-membrane charge movement (Q) in Ranvier nodes. We show that OETX reversibly blocks both sodium and displacement current. Studies of I_{Na} and Q during partial suppression by the toxin reveal differences in the effects of OETX on the remaining I_{Na} and Q. The findings are discussed in relation to recent models for the Na-channel gating process and for Na-channel block by local anaesthetics.

Oenanthe crocata is a highly poisonous plant[4,5]. The toxic substance, OETX, isolated from the roots, is a neutral ethylenic and acetylenic diol ($C_{17}H_{22}O_2$). In frog myelinated nerve fibres, OETX (0.1 mM) blocks the action potential and the sodium, potassium and displacement currents (present results and J.M.D., unpublished observations). The present experiments were carried out on voltage-clamped myelinated nerve fibres from the frog *Rana esculenta*. K current was abolished by adding tetraethylammonium (10 mM) to the external solution and by replacing the axoplasmic K content by caesium. Linear capacity and leakage currents were analogically subtracted from the total current[6]. Charge movement was recorded in the presence of tetrodotoxin (TTX, 10^{-6} M). To minimize noise and to obtain direct estimates of charge transfer, the current remaining after subtracting linear components was analogically integrated and then digitized and averaged. Typical recordings of charge transfer (integral of the asymmetrical capacity current) are presented in Fig. 1. In agreement with earlier reports[2,7], charge transfer followed a single exponential time course for moderate depolarizations ($E \lesssim 0$ mV). For larger depolarizations, two exponentials were required to describe the charge transfer kinetics.

External application of 60 μM OETX reversibly reduced Q (Fig. 1) and I_{Na} (Fig. 2) to 5–10% of their original values. These

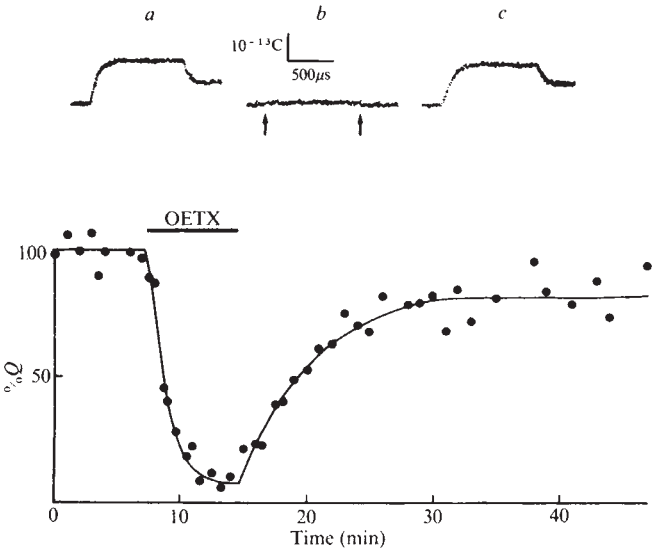

Fig. 1 Reversible immobilization of intra-membrane charges by OETX. The averaged integrated gating current was obtained in response to 12 100-mV depolarizing and 24 50-mV hyperpolarizing pulses from a holding potential of −100 mV. Temperature, 12 °C. Upper: *a*, in the control solution; *b*, in the presence of 60 μM OETX; *c*, after a 24-min wash with the control solution. Lower: relative amplitude of the charge transfer (Q) before, during and after the application of OETX (60 μM). The amplitude of Q was normalized to its mean value before the application of the toxin. Absolute charge and current amplitudes were in all cases calculated assuming a 10-MΩ internal resistance. A 4×10^{-2} solution of OETX in ethanol was diluted in Ringer's solution to give final toxin concentrations. Fibre 6-11-79 A.

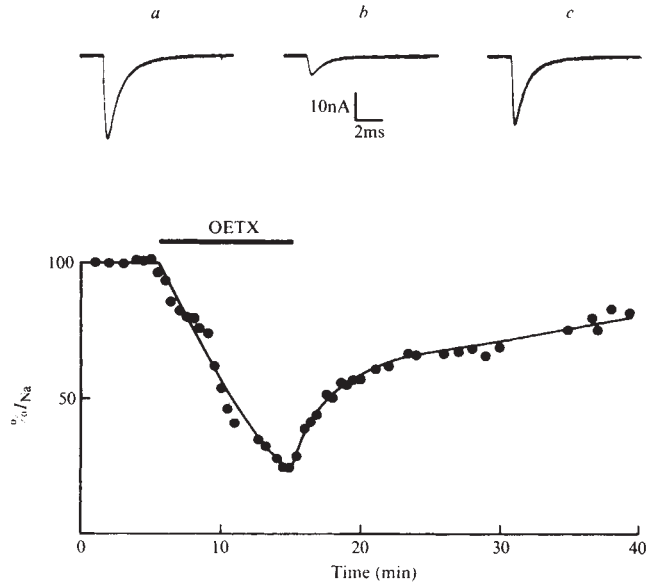

Fig. 2 Reversible block of I_{Na} by OETX. Sodium current was recorded at $E = 0$ mV after a 50-ms prepulse to −110 mV (control) or −140 mV (OETX). Holding potential, −70 mV. Temperature, 12 °C. Upper: *a*, in the control solution; *b*, in the presence of 60 μM OETX; *c*, after a 23-min wash with control solution. Lower: relative amplitude of peak I_{Na} before, during and after the application of OETX (60 μM). The amplitude of I_{Na} was normalized to its mean value before the application of the toxin. Fibre 13-11-79.

effects cannot be attributed to either fast (Fig. 3*c*) or slow[8] inactivation because they could not be removed by hyperpolarized holding potentials. The reduction of I_{Na} and Q were both independent of the frequency of stimulation. Recordings of sodium current and charge transfer obtained by intermittent application of saxitoxin during the development of OETX block indicated that OETX decreased I_{Na} and Q in parallel[9]. The leakage and the symmetrical capacity currents remained unchanged when I_{Na} and Q were virtually completely blocked by OETX (60 μM)[9].

In addition to reducing I_{Na} and Q, OETX altered various properties of non-blocked Na channels. The fractional depression of peak I_{Na} produced by OETX varied with voltage (Fig. 3*a*), indicating that the Na-permeability activation curve was decreased in slope with a shift of its 50% point towards positive voltages (Fig. 3*b*). Similar results were obtained in four other fibres in which this point was investigated. In contrast, the position and relative steepness of the Q_{on}-voltage curve were not significantly modified by toxin treatment which reduced Q_{on} at 0 mV to 34–57% of control in three fibres (Fig. 4*a*). This is shown by Fig. 4*b*, in which the Q_{on} results in OETX are replotted with values for each fibre scaled up so as to compensate for the depression of Q by OETX. Both the data points in Fig. 4*b* and their best fit (dashed curve) by the two-state Boltzmann model (Fig. 4 legend) lie close to the fit (solid curve) to the control Q_{on} data from the same fibres which are reproduced from Fig. 4*a* in 4*b*. The effect of OETX on the Na-permeability activation curve cannot be attributed to a decrease in the series resistance artefact due to the smaller I_{Na} (ref. 10) because a reduction of I_{Na} to 40% of its original value by TTX (4×10^{-9} M) caused the Na-permeability activation curve to be shifted towards positive voltages by less than 2 mV (refs 9, 11). The time course of Na-current activation, determined after correction for a two-exponential inactivation process[12] in parallel, was either slightly delayed or not significantly affected by OETX[9], whereas the time constants for charge movement were reduced[9].

OETX markedly shifted the Na-inactivation curve towards negative potentials and slightly decreased its slope (Fig. 3*c*). The

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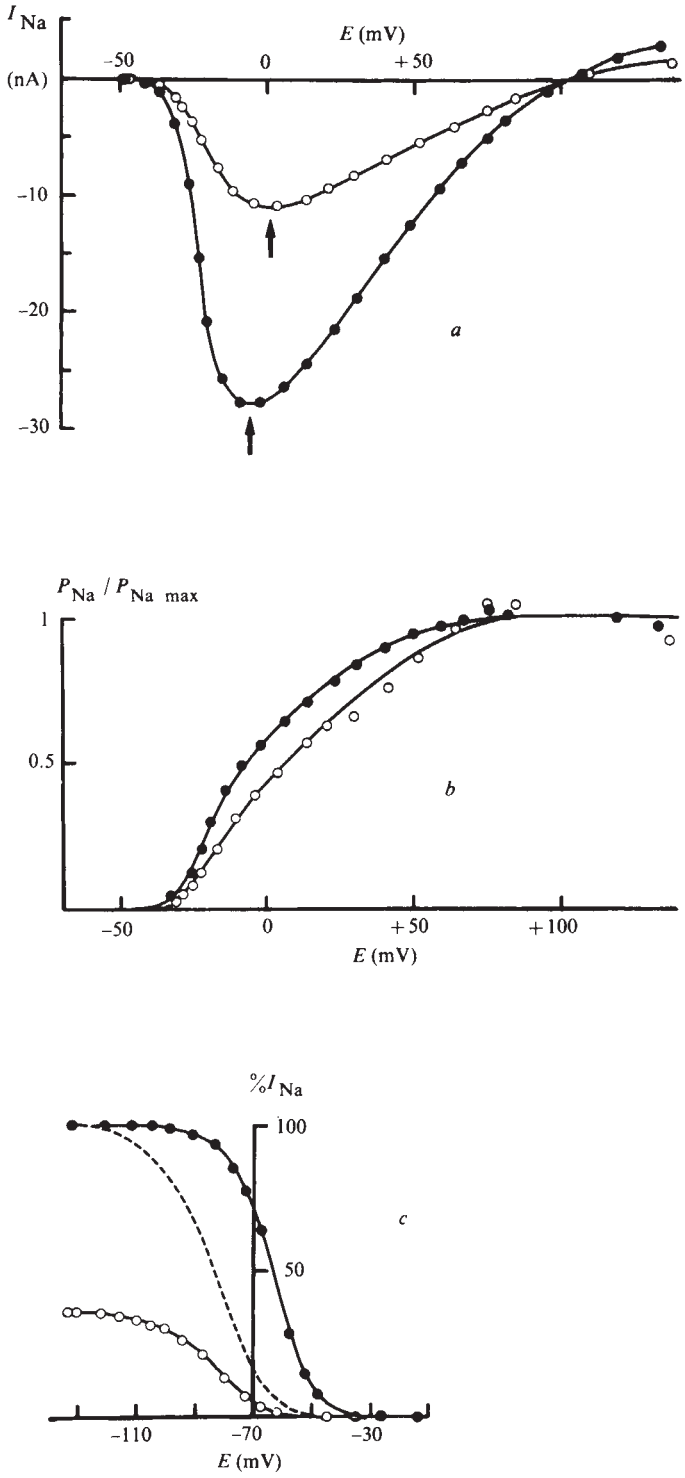

Fig. 3 Na current (*a*), Na permeability (*b*) and Na-current inactivation (*c*) voltage curves in control solution (filled circles) and during wash shortly after exposure to OETX (200 μM) (open circles). Holding potential, −70 mV. Temperature, 12 °C. *a*, The peak of I_{Na} was recorded during test pulses preceded by 50-ms prepulses to −110 mV (control solution) or −140 mV (during wash), so as to ensure complete removal of Na inactivation. Note that under the effect of toxin, the potential at which the inward current is maximum (arrows) is shifted towards positive values. *b*, Relative peak Na permeability (P_{Na}) as a function of membrane potential calculated from each set of results in *a* assuming the Na channel to rectify according to the constant field equation[19]. *c*, Available Na current (per cent of maximum control I_{Na}) as a function of membrane potential during a 50-ms prepulse. The dashed curve gives the results in OETX normalized to a maximum value of 100% for large hyperpolarizations. I_{Na} was measured at 0 mV. Fibre 26-2-79.

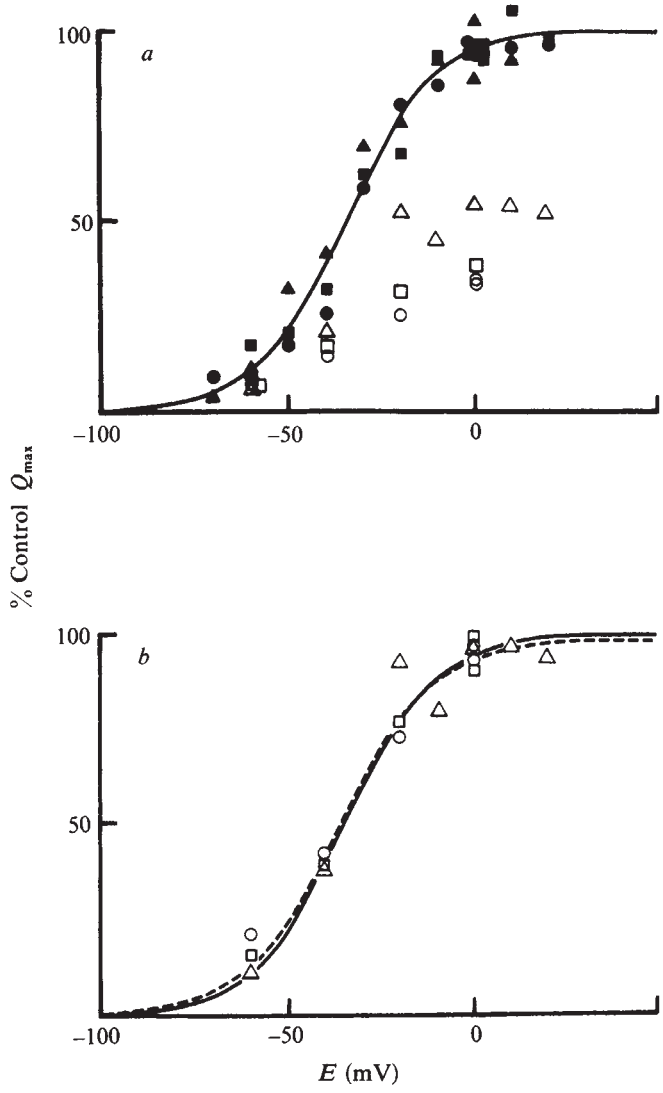

Fig. 4 Relative charge–voltage relationship in control solution (filled symbols) and in the presence of 40–60 μM OETX (open symbols). Holding potential, −100 mV. Temperature, 10–12 °C. *a*, Values obtained in both solutions on each of three fibres (different symbols) were first normalized to the mean control value of Q_{on} in that fibre at 0 mV. A non-linear least squares fit of the equation

$$Q_{on} = Q_{max}/\{1 + \exp[(\bar{E} - E)/k]\} \quad (1)$$

for the two-state Boltzman model[20–22] was first made to the combined values from the three fibres in control solution. All Q_{on} values were then normalized to the control Q_{max}. The values of Q_{max} (normalized), $\bar{E}$ and k obtained from the fit (curve) were 100%, −34.3 mV and 12.0 mV, respectively. *b*, Q values in OETX for each fibre were scaled by a constant chosen so as to minimize the squared differences between scaled values and the fit to the combined control data in *a*. The fractional values of Q in OETX determined from these scale factors were 35% (○), 57% (△) and 40% (□) of the control fit. Equation (1) was then fitted to the combined scaled OETX results, giving the dashed curve (Q_{max} = 98.1%, $\bar{E}$ = −36.4 mV and k = 12.6 mV). The fit to the control data (solid curve) is reproduced from *a* for comparison. Fibres 15-10-79 (○), 6-11-79A (△), 21-12-79 (□).

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kinetics of Na inactivation were accelerated[9] and the ratio Q_{off}/Q_{on} for a given pulse duration was slightly decreased.

Two major conclusions can be drawn from these results: OETX induces a reversible block of I_{Na} and charge movement, and in the presence of the toxin, the voltage and time dependences of the remaining sodium current and charge movement seem to be altered differently. Because the latter conclusion is based on measurements of OETX effects in different fibres with probable differences in toxin exposure, a quantitative comparison of toxin effects on I_{Na} and Q was not attempted.

In some respects, the effects of the uncharged toxin OETX seem to resemble those of the uncharged local anaesthetic benzocaine, which depresses both I_{Na} (refs 8, 13) and Q (ref. 14) and shifts and decreases the slope of the Na-inactivation curve[13]. However, whereas maximum attainable concentrations of benzocaine produce only partial depression of sodium current[8,15,16] and charge movement (ref. 14 and our unpublished observations), OETX can almost completely suppress I_{Na} and Q. This difference may be due to an inability to achieve sufficiently high benzocaine levels at the reactive channel site for all channels to react with drug, whereas OETX is sufficiently more soluble in the membrane or more reactive with the site to occupy almost all the sites.

In view of the similarities between the effects of OETX and benzocaine, it seems likely that the OETX effects on steady-state parameters may be accounted for by a general scheme for local anaesthetic action in which the drug can bind to the Na channel in each of its several states[13]. The depression of I_{Na} which cannot be restored by hyperpolarization would be due to OETX binding to resting channels. The shift and change in slope of the Na-inactivation curve would be explained by drug binding to open and inactivated channels. With this interpretation, the depression of Q would indicate that drug-blocked channels cannot undergo transitions between states, at least not transitions giving rise to detectable charge movement. Such a scheme for OETX action may perhaps also account for the modification of kinetic parameters. Alternatively, in addition to binding to the channel in its various states, it is possible that OETX produces the observed kinetic effects by acting at a different site from that at which it modifies the rate constants for channel transitions. In either case, the possibility of multiple transitions between resting and activated states[2,17], which may contribute differing amounts of Q, may perhaps account for the differences observed between OETX effects on the kinetics of I_{Na} and Q and on the voltage dependencies of sodium permeability and Q. The accelerated Na inactivation and decreased Q_{off}/Q_{on} are consistent with the suggested relationship between inactivation and charge immobilization[2,18].

We thank J. Louvel and Y. Chauvel for providing OETX samples and N. L. Sizto for running the curve fitting program. M.F.S. was on leave from the Department of Physiology, University of Rochester, Rochester, New York 14642.

Received 23 June; accepted 2 December 1980.

1. Almers, W. *Rev. Physiol. Biochem. Pharmac.* **82**, 96–190 (1978).
2. Armstrong, C. M. & Bezanilla, F. *J. gen. Physiol.* **70**, 567–590 (1977).
3. Anger, J. P. *et al. Eur. J. Tox.* **9**, 119–125 (1976).
4. Grundy, H. F. & Howarth, F. *Br. J. Pharmac.* **11**, 225–230 (1956).
5. Chauvel, P., Louvel, J., Anger, J. P. & Chauvel, Y. *C. r. hebd. Acad. Sci. Séance, Paris* **286**, 1833–1836 (1978).
6. Dubois, J. M. & Bergman, C. *Pflügers Arch. ges. Physiol.* **370**, 185–194 (1977).
7. Nonner, W., Rojas, E. & Stämpfli, R. *Pflügers Arch. ges. Physiol.* **375**, 75–85 (1978).
8. Khodorov, B., Shishkova, L., Peganov, E. & Revenko, S. *Biochim. biophys. Acta* **433**, 409–435 (1976).
9. Dubois, J. M. & Schneider, M. F. *Adv. physiol. Sci.* (in the press).
10. Drouin, H. & Neumcke, B. *Pflügers Arch. ges. Physiol.* **351**, 207–229 (1974).
11. Neumcke, B., Nonner, W. & Stämpfli, R. *Pflügers Arch. ges. Physiol.* **363**, 193–203 (1976).
12. Chiu, S. Y. *J. Physiol., Lond.* **273**, 573–596 (1977).
13. Hille, B. *J. gen. Physiol.* **69**, 497–515 (1977).
14. Khodorov, B., Guselnikova, G. & Peganov, E. *Dokl. Acad. Nauk SSSR* **244**, 1252–1255 (1979).
15. Arhem, P. & Frankenhaeuser, B. *Acta physiol. scand.* **91**, 11–21 (1974).
16. Hille, B. *J. gen. Physiol.* **69**, 475–496 (1977).
17. Armstrong, C. M. & Gilly, W. F. *J. gen. Physiol.* **74**, 691–711 (1979).
18. Nonner, W. *J. Physiol., Lond.* **299**, 573–603 (1980).
19. Dodge, F. A. & Frankenhaeuser, B. *J. Physiol., Lond.* **148**, 188–200 (1959).
20. Schneider, M. F. & Chandler, W. K. *Nature* **242**, 244–246 (1973).
21. Keynes, R. D. & Rojas, E. *J. Physiol., Lond.* **239**, 393–434 (1974).
22. Nonner, W., Rojas, E. & Stämpfli, R. *Pflügers Arch. ges. Physiol.* **354**, 1–18 (1975).

Fibrinogen is the receptor for the endogenous lectin of human platelets

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Washed platelets activated by α-thrombin, γ-thrombin[1], thrombocytin[2] or the ionophore A23187 (ref. 3) lose their disk shape[4], produce pseudopodia and become cohesive. This cohesiveness is accompanied by the expression of an endogeneous haemagglutinin[5] which, although apparently bound to the platelet membrane[6], is dependent on cell secretion[7]. The interaction of this agglutinin with appropriate receptors on other platelets is believed to be responsible for aggregation. We report here that platelets can be prepared which lack agglutinin activity but have receptor function, that afibrinogenaemic platelets lack receptor activity, and that fibrinogen is the receptor for the agglutinin secreted by activated platelets.

Washed, fixed and non-fixed platelets were examined for thrombin-enhanced agglutinin activity using microtitre plates (Table 1). It is known that non-activated platelets do not express the agglutinin[5], and accordingly non-activated washed platelets did not agglutinate either themselves or fixed platelets. In contrast, platelets activated by thrombin agglutinated themselves and their agglutination titre was increased two- to fourfold by the presence of fixed platelets. The formaldehyde-fixed platelets, when treated with thrombin, neither agglutinated themselves nor fixed bovine erythrocytes which had receptors for the platelet agglutinin. Thus, the formaldehyde-fixed platelets had no agglutinin activity, which means that the apparent increase in the agglutination titre resulting from the presence of the fixed platelets was due to agglutination of the latter by the agglutinin of the thrombin-activated washed platelets. These results demonstrate that fixed platelets have functional receptors for the agglutinin but no functional agglutinin and that they did not cause activation of the washed platelets.

Agglutination of the fixed platelets by the activated washed platelets in the microtitre plate assays was blocked by the same compounds which block thrombin-induced platelet aggregation and enhanced haemagglutination[5] (see Table 2). Amino sugars and arginine completely abolished this activity whereas neutral sugars and other amino acids had no effect. Therefore, the binding specificity of the receptor on fixed platelets seems to be indistinguishable from that of either washed platelets or platelets in plasma[5].

In the course of characterizing platelets from patients with bleeding disorders, it was discovered that fixed platelets from a patient with congenital afibrinogenaemia[8] failed to enhance the agglutination titre of thrombin-activated platelets, which means that the fixed afibrinogenaemic platelets lacked agglutinin receptor function (Table 3). Furthermore, it was observed that this deficiency could be corrected if the afibrinogenaemic platelet-rich plasma was diluted 1:1, before fixation, with platelet-poor plasma from a donor who had a normal level of plasma fibrinogen. These results, and evidence that platelets from the afibrinogenaemic patient have no obvious plasma membrane deficiencies (D. R. Phillips, personal communication), led us to speculate that fibrinogen is the receptor for the secreted agglutinin expressed by activated platelets.

This hypothesis was tested by determining if fibrinogen could inhibit the expression of the enhanced haemagglutinin activity of activated platelets, the rationale being that free fibrinogen

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Table 1 Fixed platelets lack agglutinin activity in microtitre plate assays

Diluted material	Fixed cells added	Titre
Washed platelets	None	0
Thrombin-activated washed platelets	None	2
Thrombin-activated washed platelets	Bovine erythrocytes	16*
Fixed platelets	None	0
Fixed platelets plus thrombin	None	0
Fixed platelets plus thrombin	Bovine erythrocytes	0

Agglutination of platelets was assayed in microtitre 'V' plates (Cooke Engineering Laboratory Products)[6]. 25 μl of Tris-buffered saline containing calcium (TCS buffer: 10 mM Tris, 2 mM $CaCl_2$, 150 mM NaCl, *p*H 7.4) was added to each well then 25 μl of the diluted material was added to the first in a row of eight wells and was serially diluted (×2) in a series of six wells. The remaining two wells contained no diluted materials and served as controls. Finally, 25 μl of the fixed cells added were placed into each well, as either a suspension of fixed platelets (1×10^8 ml^{-1}) or a 2.5% suspension of trypsin-treated, formalinized bovine erythrocytes in TCS buffer[6]. Wells not containing fixed cells received 25 μl of TCS buffer and the contents of the wells were agitated gently. Each test was run in triplicate. Well contents were evaluated for agglutination after standing for 7 h at room temperature. The titre of a diluted material is defined as the reciprocal of the end point dilution (greatest dilution that gives agglutination). The platelets used as diluted materials were suspended in Tris-buffered saline containing EDTA (ETS buffer: 1 mM EDTA, 10 mM Tris, 150 mM NaCl, 0.2% α-D-glucose, *p*H 7.4) at a concentration of 10^9 ml^{-1}. Washed and fixed platelets were treated with thrombin by adding a final concentration of 0.25 U ml^{-1} of human thrombin (provided by Dr J. Fenton II) to the platelets, incubating the mixture for 1 min at 22 °C, adding it to the well and diluting immediately. Platelets were fixed by making a 1 : 1 dilution of platelet-rich plasma, obtained from blood drawn into ACD (2.5 g Na_3 Citrate. $2H_2O$, 2 g glucose, 1.5 g citric acid in 100 ml H_2O), into modified phosphate-buffered saline (40 mM PO_4, 150 mM NaCl, *p*H adjusted to 7.4 with NaOH) containing 0.5% formaldehyde and incubating overnight with gentle shaking. All operations were done at 37 °C. The fixed platelets were washed three times with TS buffer (10 mM Tris, 150 mM NaCl, *p*H 7.4) and suspended in TCS before use. Fixed platelets were used on the same day as they were washed. Suspensions of fixed non-activated platelets were birefringent, and the platelets had a disk shape when viewed in a scanning electron microscope. ADP treatment of the platelets (final concentration 5 μM ADP) before fixation typically decreased their receptor function—this may be caused by the generation of excess receptor activity but has not been tested.

* This agglutination was inhibited by a 30 mM final concentration of each of the following: L-arginine, α-D-mannosamine, α-D-glucosamine, and α-D-galactosamine but not by a final concentration of 60 mM glutamine, *N*-acetylglucosamine or *N*-acetylgalactosamine.

Table 2 Fixed platelets have receptor activity for the thrombin-enhanced agglutinin

Diluted material	Fixed cells added	Titre
Washed platelets	None	0
Washed platelets	Platelets	0
Thrombin-activated washed platelets	None	2
Thrombin-activated washed platelets	Platelets	8*

Assays were performed as described in Table 1 legend.

* This agglutination was inhibited by the same compounds which inhibit thrombin-induced platelet aggregation and the function of the platelet-mediated enhanced haemagglutination activity of thrombin-activated washed platelets[5], that is, by a 30 mM final concentration of each of the following: L-arginine, α-D-mannosamine, α-D-glucosamine, and α-D-galactosamine but not by a final concentration of 69 mM glutamine, *N*-acetylglucosamine or *N*-acetylgalactosamine.

could compete with cells for the secreted agglutinin, if it functions as an agglutinin receptor. Washed platelets activated with α-thrombin, γ-thrombin, thrombocytin or A23187 were tested for enchanced haemagglutination activity in the presence and absence of fibrinogen. As shown in Fig. 1, fibrinogen completely inhibited the expression of the agglutinin in the absence of Ca^{2+} but had no effect in the presence of Ca^{2+}. The fact that fibrinogen did not inhibit the function of the agglutinin in the presence of Ca^{2+} explains why plasma fibrinogen would not be expected to inhibit the function of the endogenous platelet agglutinin *in vivo*. The inhibitory effect seems to be specific for fibrinogen because neither albumin nor cold insoluble globulin caused inhibition. These results may partially explain the observation that monovalent anti-fibrinogen antibody fragments drastically inhibited the aggregation of thrombin-activated washed platelets[9]. The basis of the ability of Ca^{2+} to eliminate the fibrinogen-induced inhibition of enhanced

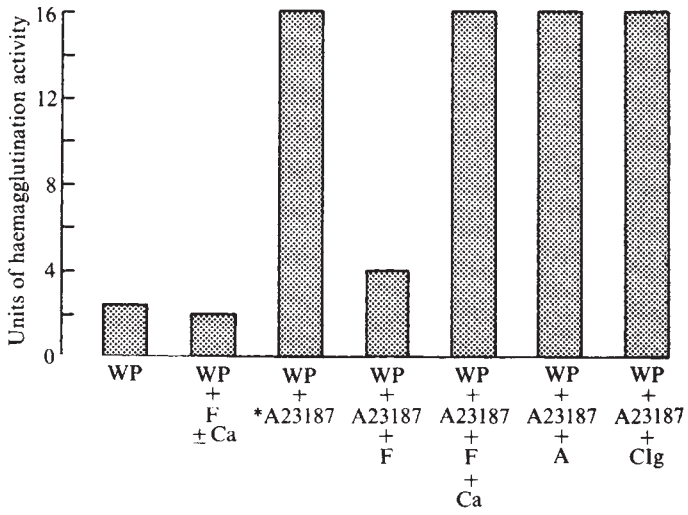

Fig. 1 Microtitre plate assays were carried out as described in Table 1. TS was the plate buffer and calcium ($CaCl_2$) was added where indicated at a final concentration of 2 mM. Platelets suspended in ETS were activated with one of the following: A23187 at 1 μg ml^{-1} (Calbiochem), α-thrombin at 0.25 U ml^{-1}, γ-thrombin at 5 μg ml^{-1} (both provided by Dr J. Fenton II), and thrombocytin at 5 μg ml^{-1} (supplied by Dr K. Stocker, Pentapharm). Human fibrinogen (L-grade, Kabi) treated with diisopropyl fluorophosphate[13], was used at a final concentration of 250 μg ml^{-1}. 125 μg ml^{-1} of fibrinogen caused inhibition of half the maximum haemagglutination activity. Similar results were obtained using washed platelets activated with each stimulus listed above. Cold insoluble globulin (CIg, supplied by Dr K. Yamada[14]) was used at a final concentration of 125 μg ml^{-1}. Human serum albumin (Sigma) was used at a final concentration of 250 μg ml^{-1}. WP, washed platelets; F, fibrinogen; Ca, calcium; A, albumin; CIg, cold insoluble globulin. * The haemagglutination caused by washed platelets activated by A23187 was not inhibited by the supernatant fraction from heat-precipitated fibrinogen.

haemagglutination activity is unknown, but a possible explanation is that fibrinogen bound to platelets in the presence of Ca^{2+} has a much greater affinity for the agglutinin than does free fibrinogen.

The possibility that fibrinogen is the haemagglutinin was evaluated by the following tests. Fibrinogen itself was found to have no haemagglutination activity[7] (Fig. 1). Washed platelets suspended in ETS (see Table 1 legend) or Tyrode buffer were treated with fibrinogen and ADP (platelets in Tyrode buffer aggregated in response to stirring in the test conditions) and found to have no enhanced haemagglutination activity. Thus there is no evidence that plasma fibrinogen is the agglutinin, but our results do not exclude the possibility that platelet fibrinogen is the agglutinin. However, as there is evidence that platelet and plasma fibrinogen are identical[10], this possibility will not be considered further. The observations that amino sugars inhibit the binding of fibrinogen to ADP-treated platelets[11] does not elucidate whether fibrinogen functions as an agglutinin rather than a receptor for the agglutinin, but it does explain how these compounds can inhibit platelet aggregation[5].

Table 3 Agglutinin receptor activity of formaldehyde-fixed afibrinogenaemic platelets

Diluted material	Fixed cells added	Titre
Washed platelets	None	0
Thrombin-activated washed platelets	None	2
Thrombin-activated washed platelets	Control platelets	8
Thrombin-activated washed platelets	Afibrinogenaemic platelets	2
Thrombin-activated washed platelets	Afribinogenaemic platelets fixed in diluted control platelet-poor plasma	8

Assays were performed as described in Table 1 legend. The patient with congenital afibrinogenaemia had <10 μg ml^{-1} of plasma fibrinogen and <0.6 μg fibrinogen per 10^8 platelets.

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The results described here demonstrate that the agglutinin of activated platelets is distinct from its receptor and that fibrinogen is the receptor for the agglutinin. According to these results and those already published[5,7], secreted materials, including proteins, mediate platelet aggregation by cross-linking adjacent platelets. This interaction of secreted agglutinin with platelets is mediated by fibrinogen—that is, fibrinogen links the agglutinin to the platelet plasma membranes. Fibrinogen, in turn, is probably specifically bound to the plasma membranes via membrane glycoproteins. The identity of these glycoproteins has not been reported, but evidence suggests that platelet glycoproteins IIb and/or III may be the fibrinogen receptors[12].

This work was supported by USPHS grants HL 23010 and HL 11880.

Received 30 June; accepted 9 December 1980.

1. Fenton, J. W. II, Landis, B. H., Walz, D. A. & Finlayson, J. S. in *Chemistry and Biology of Thrombin* (eds Lundblad, R. L., Fenton, J. W. II & Mann, K. G.) 43–70 (Ann Arbor Science, Michigan, 1977).
2. Kirby, E. P. *et al. Biochemistry* **18**, 3564–3570 (1979).
3. Feinman, R. D. & Detwiler, T. C. *Nature* **249**, 172–173 (1974).
4. White, J. G. *Am. J. Path.* **71**, 363–378 (1979).
5. Gartner, T. K., Williams, D. C., Minion, F. C. & Phillips, D. R. *Science* **200**, 1281–1283 (1978).
6. Gartner, T. K., Williams, D. C. & Phillips, D. R. *Biochem. biophys. Res. Commun.* **79**, 592–599 (1977).
7. Gartner, T. K., Phillips, D. R. & Williams, D. C. *FEBS Lett.* **113**, 196–200 (1980).
8. Shapiro, R. S. *et al. Am. J. Pediat. Hemat. Oncol.* **2**, 207–212 (1980).
9. Tollefsen, D. M. & Majerus, P. W. *J. Clin. Invest.* **55**, 1259–1268 (1975).
10. Doolittle, R. F., Takagi, T. & Cottrell, B. A. *Science* **185**, 368–370 (1974).
11. Kinlough-Rathbone, R. L., Chahil, A., Perry, D. W., Packham, M. A. & Mustard, J. F. *Blood* **54**, 249 (1979).
12. Bennett, J. S. & Vilaire, G. *J. clin. Invest.* **64**, 1393–1401 (1979).
13. Mustard, J. F., Perry, W., Kinlough-Rathbone, R. L & Packham, M. A. *Am. J. Physiol.* **228**, 1757–1765 (1975).
14. Yamada, K. M. & Kennedy, D. W. *J. Cell Biol.* **80**, 492–498 (1979).

Non-uniform ion distributions and electrical potentials in sarcoplasmic regions of skeletal muscle fibres

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We report here electrophysiological and ion distribution results obtained with mechanically skinned skeletal muscle fibres which indicate that a Donnan equilibrium, characterized by large ionic concentration and electrical potential differences, is established between the myofibrillar space and the rest of the sarcoplasm surrounding the myofibrils. The more negative electrical potential within the myofibrils (−15 to −20 mV for conditions similar to those *in vivo*) can strongly influence: (1) intracellular Ca^{2+} movements associated with contraction; (2) the location of sarcoplasmic enzymes, their substrates and products of reaction; (3) measurements of the electrical potential difference between the extra- and intracellular space and estimation of relative membrane permeabilities for certain ions.

We used mechanically skinned single muscle fibres of the barnacle[1] (*Balanus nigrescens*), subsequently treated with a nonionic detergent which disrupted and removed all cellular membranes. Electron microscopic observations confirmed this and showed that the only regions which could be distinguished in the cross-section of the preparations after this procedure were the myofibrils and the inter-myofibrillar space.

Potential differences were measured between two glass micro-electrodes impaled into the skinned muscle fibre. As can be seen in Fig. 1, one could record either a very small or a relatively large potential difference regardless of whether both electrodes were in the preparation (Fig. 1*A*, *D*), or only one electrode was impaled in the skinned fibre while the reference electrode was kept in the surrounding solution (Fig. 1*B*). The results indicate that one of the regions within these skinned muscle fibres had essentially the same electrical potential as the surrounding solution, but that a second region yielded negative potentials of large amplitude relative to the bathing solution. When using low-resistance microelectrodes with an external tip similar in size to the diameter of myofibrils (2–3 μm), we were unable to record the large potential differences. This suggests that the former region may be associated with the space around myofibrils, whereas the latter may represent the myofibrils themselves.

The potential differences measured in these skinned muscle fibres did not change following the partial replacement of K^+

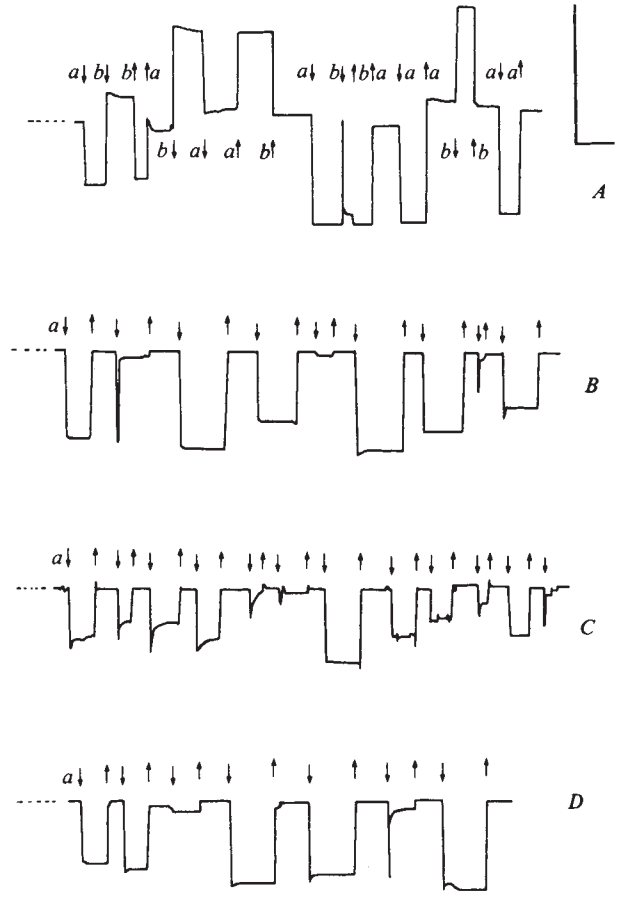

Fig. 1 Potential differences within mechanically skinned muscle fibres recorded between two glass microelectrodes denoted *a* and *b*. In *A–C* the fibres were incubated in 2% v/v Triton X-100 for 12 h after being mechanically skinned. Electrode advancement and withdrawal are indicated by ↓ and ↑, respectively. The potential difference between electrodes when both were in bathing solution is shown at the beginning of each trace by the dotted line. *A*, Records obtained with both electrodes inside a barnacle muscle fibre. In this case ↑ and ↓ refer to electrode movements within the fibre. *B*, Potentials recorded between the bathing solution (electrode *b*) and a barnacle fibre (electrode *a*). ↓ Indicates fibre impalement and ↑ withdrawal of the electrode into the bathing solution. *C*, Potential differences recorded from a toad iliofibularis fibre as in *B*. *D*, Records from a freshly skinned barnacle muscle fibre immersed in oil. In this instance the dotted line represents the potential difference between electrodes when just in contact with the fibre surface. The internal resistance of the glass microelectrodes (normally 40–80 MΩ when filled with either 2 M KCl or 4 M Na-acetate) was routinely measured when the electrodes were in solution and in the preparation to detect and discard artefactual potential measurements due to electrode tip blockage and/or damage. The external tip diameters and tip potentials of these electrodes were characteristically 0.1–0.3 μm and <5 mV, respectively. Input impedance $>10^{13}$ Ω. Composition of bathing solution for *A–C* (mM): EGTA, 10; Na_2ATP, 3; $MgCl_2$, 2; K^+, 100; trimethylaminoethanesulphonic acid (TES), 40; *p*H 7.10 adjusted with Cl. Calibration bars: vertical 30 mV; horizontal 30 s.

0028–0836/81/080690—03$01.00
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Table 1 R_i values for different substances obtained in skinned muscle preparations from nine barnacles in different ionic conditions and predicted potential differences (ΔV^i_{pred}) between the myofibrillar and extra-myofibrillar space

$\Gamma/2$ (mM)*	R_K	R_{EGTA}	R_{Gly}	R_{BSA}	R_{Carb}	R_{Mk}	R_{Myo}	R_{Chym}	$R_{Cyt\,c}$	R_{Aldo}	ΔV^K_{pred} (mV)†	ΔV^{EGTA}_{pred} (mV)†
55	1.62±0.03	0.53±0.02	0.99±0.01	0.53±0.02	0.73±0.04	—	1.2±0.03	—	4.6±0.2	1.27±0.03	−22±2	<−30
80	1.45±0.02	0.59±0.02	0.98‡	0.57±0.02	—	0.77±0.02	—	—	—	—	−19±2	<−25
120	1.30±0.02	0.69±0.03	0.97±0.02	0.60±0.01	0.79±0.02	—	0.98±0.03	2.06±0.02	2.4±0.1	0.85±0.02	−16±2	−18±5
220	1.17±0.02	0.77±0.02	0.966‡	0.63±0.01	—	0.82±0.02	—	—	—	—	−12±1.5	−11±3
365	1.09±0.02	0.83±0.02	0.956‡	0.66±0.01	—	—	—	1.55±0.02	—	—	−9.5±1	−7±2
510	0.98±0.02	0.95±0.01	0.95±0.01	0.69±0.02	0.85±0.02	—	0.92±0.02	—	1.06±0.03	0.8±0.02	−2.5±2	0±1

Abbreviations: Gly, glycine; BSA, bovine serum albumin; Carb, carbonic anhydrase; Chym, α-chymotrypsinogen A; Myo, myoglobin; Aldo, aldolase; Cyt *c*, cytochrome *c*; Mk, myokinase (see ref. 3 for M_r and *p*I values). To determine R_i, a substantial mass (60–100 mg) of skinned muscle fibres (~1 g of skinned fibres could be obtained from a single barnacle) was initially equilibrated in the appropriate solution containing the substance *i* and then transferred after careful blotting to an 'unloading' solution of specially chosen composition to allow accurate determinations of the amount of *i* carried by the known mass of skinned muscle fibres into the unloading solutions. For protein determinations the unloading solutions had the composition listed in the Fig. 1 legend and for $EGTA^{2-}$ and K^+ determinations the unloading solutions contained only 200 mM KCl and 100 mM Tris-HCl (*p*H 7.10), respectively. The concentration of proteins in the loading solutions was 10 mg ml^{-1}. This concentration of proteins did not have any significant effect on R_K, R_{EGTA} or ΔV measurements. Similar results were obtained in control experiments in which the concentration of BSA was varied between 1 and 50 mg ml^{-1}. The density of the fibres was also determined in various solutions by dividing the mass of the fibres by the volume they displaced in these solutions. The initial mass of the muscle preparations mechanically skinned under paraffin oil increased by 40–50% following the detergent treatment and equilibration in solutions containing 100 mM K^+. Their mass increased further after equilibration in solutions with a higher ionic concentration[9]. The following analytical methods were used for the various R_i determinations: R_{EGTA}, titration method of Moisescu and Pusch[10]; R_{Gly}, spectrophotometric method using glycine reaction with ninhydrin; $R_{protein}$, trichloracetic acid precipitation followed by Lowry's method for protein determinations: $R_{Cyt\,c}$, both Lowry's method and direct spectrophotometric measurements; R_K, atomic absorption spectrophotometry.

* Composition of solutions as in Fig. 2 legend.

† $RT/F = 25.5$ mV.

‡ Interpolated values.

with Na^+, indicating the absence of a selective barrier, such as a membrane, between the two regions. An increase in $[Ca^{2+}]$ from ~10^{-8} M to 10^{-5} M also did not result in any significant change in the potential differences recorded. However, an increase in $[Mg^{2+}]$ from ~0.2 to 5 mM resulted in a significant decrease in the potential difference between the two regions (~5 mV for solutions similar to those in Fig. 1). Relatively large potential differences could also be recorded in mechanically skinned amphibian muscle fibres (Fig. 1*C*) and in freshly skinned barnacle muscle fibres immersed in oil (Fig. 1*D*).

To characterize further the potential difference (ΔV) between the myofibrillar space and extra-myofibrillar space, we investigated the effect on ΔV of varying the concentration of KCl in the bathing solution. As can be seen from Fig. 2, ΔV decreases with an increase in the ionic concentration of the solutions in a fashion similar to that expected for a Donnan potential[2].

An important inference from the existence of a Donnan equilibrium between different regions of the muscle fibre is the non-uniform distribution of ions in this cell, and we have used the same skinned muscle preparation to determine the distribution of ions between the myofibrillar and extra-myofibrillar space. The extra-myofibrillar region of the skinned muscle fibre can be considered as an extension of the space surrounding the preparation which is in electrochemical (Donnan) equilibrium with the space between myofilaments. If there was an important difference in the ionic composition between the myofibrillar and extra-myofibrillar space due to a Donnan-type equilibrium produced by negatively charged myofilaments, the ratio, R_i of the average concentration of an ion *i* in the preparation to the concentration of the same ion in the bathing solution, would be expected to be significantly higher for cations than for anions (see Table 1 legend for methods used to determine R_i). This is confirmed by the results shown in Fig. 3, where the ratios for K^+ and $EGTA^{2-}$ have been obtained simultaneously in the same solutions and the same batch of skinned fibres from a single barnacle. The average values for R_K and R_{EGTA} obtained over all experiments are given in Table 1.

Figure 3 and Table 1 also show the R_i values for glycine, a small uncharged molecule, which would be expected to penetrate freely the space between myofilaments. The value of R_{Gly} can be used as an index of the fraction of total fibre volume available for diffusion (S_d), and these results indicate that most of the space within our preparations is free for diffusion in all the ionic conditions used. The fraction of total fibre volume which is extra-myofibrillar (S_e) can be estimated directly from the value of R_i of a relatively large, negatively charged protein which cannot enter the myofibrillar space and we have used bovine serum albumin (BSA, M_r 68,000, *p*I 5.89)[3] as a marker for this purpose. The values of R_{BSA} are shown in Table 1. If R_{BSA} is equated to S_e, it can be seen that the extra-myofibrillar space would represent more than 50% of the total volume of these skinned muscle preparations. Indeed, electron micrographs also indicate that large areas of extra-myofibrillar space exist in extensive parts of these Triton-treated skinned muscle fibres.

If the ions are in electrochemical equilibrium in the preparation, one can predict an average value for the electrical potential difference (ΔV^i_{pred}) between the myofibrillar and extra-myofibrillar space from the Nernst equation for each ion *i*. Using the experimentally determined values for R_i and assuming, for simplicity, that $R_{Gly} = S_d$, $R_{BSA} = S_e$ and the activity coefficients of the respective ions are similar in the myofibrillar

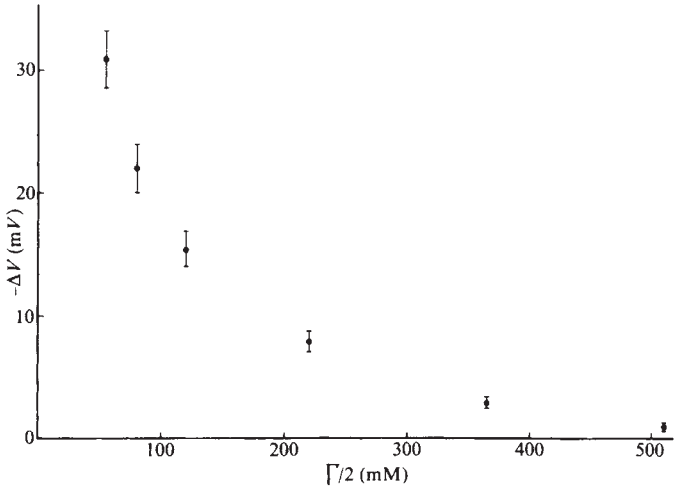

Fig. 2 Effect of ionic strength ($\Gamma/2$) on the average potential differences measured in mechanically skinned muscle fibres of the barnacle. The points represent the mean values (±s.e.m.) obtained from fibres of six different barnacles. Each fibre was impaled at least 30 times in each solution and care was taken to distribute the impalements over many regions of the fibre. The ionic strength was varied by adding differing amounts of KCl to a basic relaxing solution containing (mM): TES, 40; K_2EGTA, 10; Na_2ATP, 3; $MgCl_2$, 2. The final *p*H was adjusted with KOH to 7.10 in all solutions.

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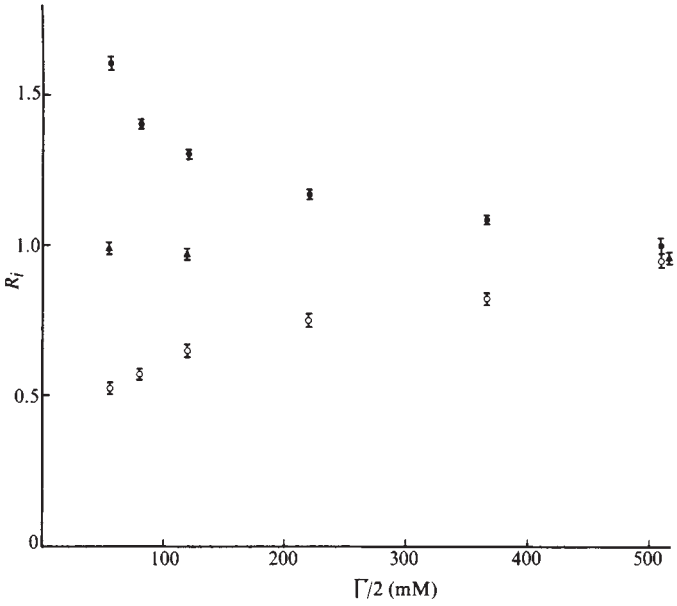

Fig. 3 Effect of ionic strength (Γ/2) on average values of R_{EGTA}(○), R_K(●) and R_{Gly}(▲), measured simultaneously in the same batch of skinned barnacle muscle fibres (200–300) from one animal. The vertical bars indicate the error of the methods (see Table 1). Note that R_{EGTA} is significantly smaller than R_K in all solutions but that of highest ionic strength and that the discrepancy between R_K and R_{EGTA} increases with decreasing ionic concentration in the solutions, as expected if a Donnan equilibrium was mainly responsible for the distribution of ions in these muscle preparations.

space and in the bulk solution, the predicted ranges for ΔV (ΔV^{K}_{pred} and ΔV^{EGTA}_{pred}) shown in Table 1 were obtained from the following equation

$$\Delta V^{i}_{pred} = (RT/n_iF) \ln (S_d - S_e)/(R_i - S_e)$$

In addition to BSA, the distribution of several other proteins of differing molecular weight and *p*I were investigated and their R_i values are listed in Table 1. The R_i values for the proteins of M_r <20,000 approach the value for R_{Gly} as the ionic strength of the solutions increases, suggesting that there was no specific binding other than an electrostatic type of interaction between filaments and cytochrome *c*, or myoglobin, and that there was no steric restriction for the diffusion of these proteins between myofilaments. Cytochrome *c* and α-chymotrypsinogen A are both highly positively charged at *p*H 7.00 and show much larger values for R_i than does K^+ in conditions where a significant potential difference exists. Myoglobin is only slightly charged in these solutions and accordingly has an R_i value smaller than R_K.

The other proteins, carbonic anhydrase, aldolase and BSA, show a significantly smaller R_i value than R_{Gly} in the presence of 500 mM K^+ compared with $R_{EGTA} \simeq R_K \simeq R_{Gly}$, suggesting that these proteins are sterically restricted to certain degrees from entering the myofibrillar space. Of these proteins only aldolase is highly positively charged and if free to diffuse between myofilaments it would have been expected to have R_i values close to $R_{cyt\,c}$, which is not the case. However, the R_i value for aldolase is significantly higher than R_{BSA} for lower ionic strength solutions and this could be due to an increased concentration of aldolase in the outer layers of myofibrils due to the negative potential differences. These results on protein distribution: (1) strongly support the existence of a significant Donnan equilibrium within muscle fibres, (2) indicate that steric restrictions can become significant for the diffusion of proteins with M_r> 20,000–30,000 and (3) suggest that the location of sarcoplasmic biochemical reactions would be determined by the charge and the size of the molecules involved. An interesting corollary from these experiments is that the prevailing electrical potential configuration within skeletal muscle fibres would promote a rapid activation of contraction by selectively facilitating the diffusion of Ca^{2+} released from the sarcoplasmic reticulum towards the area of the filament lattice with the highest negative charge density (region of filament overlap), where force is generated.

Previous attempts to record electrical potential differences between bundles of glycerinated muscle fibres[4–6] and the bathing solutions, and between mechanically skinned muscle fibres[7,8] and the bathing solutions have been reported. However, doubts have been raised about the state of membranes in the multicellular glycerinated preparations[6] and more recent observations on mechanically skinned muscle fibres indicated either a complete absence or drastically reduced potential differences[7,8] between the skinned muscle preparations and the bathing medium. The present results, obtained using several independent measurements, show that in the absence of any cellular membranes a significant electrical potential difference exists between regions within these muscle fibres and that electrically charged substances are not uniformly distributed within muscle fibres at physiological levels of monovalent cations.

We thank Mrs R. Cafarella for technical assistance and the Australian Research Grants Committee and National Heart Foundation of Australia for financial support.

Received 21 July; accepted 2 December 1980.

1. Ashley, C. C. & Moisescu, D. G. *J. Physiol., Lond.* **270**, 627–652 (1977).
2. Overbeek, J. M. G. *Prog. Biophys. molec. Biol.* **6**, 57–84 (1956).
3. Righetti, P. G. & Caravagio, T. *J. Chromat.* **127**, 1–30 (1976).
4. Collins, E. W. & Edwards, C. *Am. J. Physiol.* **221**, 1130–1133 (1971).
5. Pemrick, S. M. & Edwards, C. *J. gen. Physiol.* **64**, 551–567 (1974).
6. Weiss, R. M., Lazzara, R. & Hoffman, B. F. *Nature* **215**, 1305–1307 (1967).
7. Gordon, A. M., Godt, R. E., Donaldson, S. K. B. & Harris, C. E. *J. gen. Physiol.* **62**, 550–574 (1973).
8. Fabiato, A. & Fabiato, F. *Circulation Res.* **31**, 293–307 (1972).
9. Godt, R. E. & Maughan, D. W. *Biophys. J.* **19**, 103–116 (1977).
10. Moisescu, D. G. & Pusch, H. *Pflügers Arch. ges. Physiol.* **355**, R122 (1975).

Rhythmic contractile activity of the *in vivo* rabbit aorta

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The cellular element of the aorta is largely smooth muscle; yet this organ has long been regarded as a passive elastic tube[1–4], without regard for the possible function of its smooth muscle cells. The isolated rabbit aorta became popular as a convenient smooth muscle preparation[5], and much has been learned about its cellular physiology, but unfortunately the functional role of this tissue was lost on its excision. Like many mammalian arterial preparations, the isolated aorta either fails to show spontaneous electrical or mechanical activity, or shows activity of a much slower frequency than the rhythmic activity of the heart[6,7], thus obscuring any possible relationship with the pulsatile activity of the heart. However, contractions are elicited from the isolated aorta on stimulation with various neural and hormonal agents[5–10]. In effect, the aortic smooth muscle has been classified as multiunit, which suggests that physiological activation may be neurogenic[11]. Thus, removal of the muscle from the body may result in disruption of the neural connections which function in normal activation of contraction. We show here that when recordings of aortic tension are made *in vivo*, rhythmic contractions are observed. Evidence is presented which indicates that the contractions are neurogenic in origin and exhibit a precise phasing pattern with the pulse wave.

0028–0836/81/080692—03$01.00

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A longitudinal cut was made along the abdominal midline of rabbits anaesthetized with an intravenous injection of Nembutal (35 mg per kg) through the ear vein. The abdominal aorta was exposed, and the blood flow from a 5-cm section was bypassed using a polyethylene tube (Fig. 1). In the centre of the bypass tube, a catheter was mounted and connected to a blood pressure transducer (Statham P23Db). An isometric tension transducer (Grass FTO3C) was attached to the bypassed muscle segment such that simultaneous pressure and tension recordings could be made. These recordings were made at loci equidistant from the heart.

Illustrated in Fig. 2 are simultaneous recordings of pulse pressure, measured in the bypass tube, and muscle tension, recorded from the bypassed muscle segment. The pressure waves and muscle contractions exhibited a 1 : 1 correspondence with the phasing between the two events, displaying a definite relationship. During the pressure wave diastole, muscle tension was maximum. Generally, within 20 ms after the initiation of an increase in pressure, the muscle began to relax and tension was usually at a minimum by a time corresponding to the dichrotic notch. As pressure began to decline again, muscle tension once again increased. This increase in tension was characterized by two components, a fast and a slow phase. The relative duration of the two phases varied somewhat from preparation to preparation. Such a cyclic pattern of simultaneous pressure and tension changes was observed in 10 of 10 animals tested.

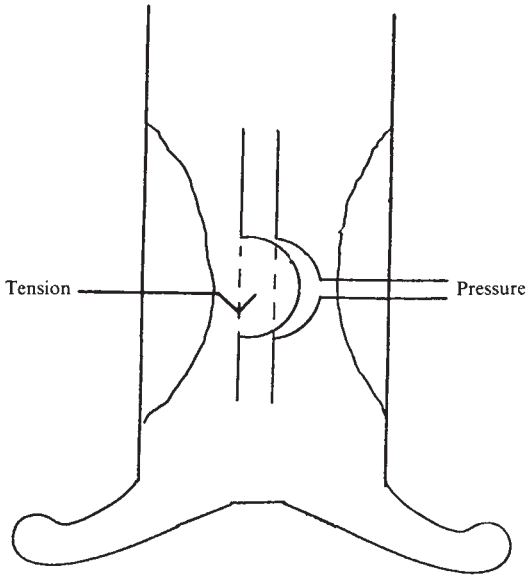

Fig. 1 Schematized recording technique for simultaneous measurement of pressure and tension in rabbit aorta. Dotted lines indicate bypassed muscle segment.

The possibility that the tension recordings were produced artefactually by body movements secondary to the pulse wave was eliminated by the lack of parallelism between the two recordings. Local application to the bypassed aortic muscle segment of the calcium flux inhibitor lanthanum (3 mM), local anaesthetics xylocaine (1%) and tetrodotoxin (10^{-7}–10^{-6} M), or the α-adrenergic blocker phentolamine (10^{-4} M) inhibited or abolished the contractions without affecting the pulse pressure. The contractions were enhanced by local application of atropine (10^{-4} M) or noradrenaline (10^{-6}–10^{-5} M), again without any effect on pulse pressure. If, on the other hand, noradrenaline (10^{-6} M) was administered parenterally through the ear vein, the pulse pressure increased while the contractions in the bypassed segment either decreased in amplitude or remained unchanged.

The abolition of the contractions by tetrodotoxin indicates that their propagation is neurally mediated. This premise was further investigated by measuring the direction and conduction velocity of the contractile wave. Figure 3 illustrates the effects of interrupting the anatomical continuity of the aorta on either side of the segment from which the contraction recordings were made. If the aorta was severed cranially (upstream), the contractions ceased completely (Fig. 3*b*), whereas after a distal cut contractions continued (Fig. 3*a*). The signal for contraction is therefore conducted in the same direction as the pulse wave. To measure the velocity of conduction, we bypassed two segments of the aorta and recorded contractions at two locations, the distance between which was accurately measured. The time interval between the onset of the contractions yielded a conduction velocity of 1.9 m s^{-1} directed caudally, a similar conduction velocity to that found for the pressure wave[12]. This velocity falls well within the range of neural activity[13,14] but is much faster than the propagation velocity observed for myogenic phenomena in other types of smooth muscle[15,16].

From the above results, we conclude that neural impulses activate waves of smooth muscle contraction, travelling with the

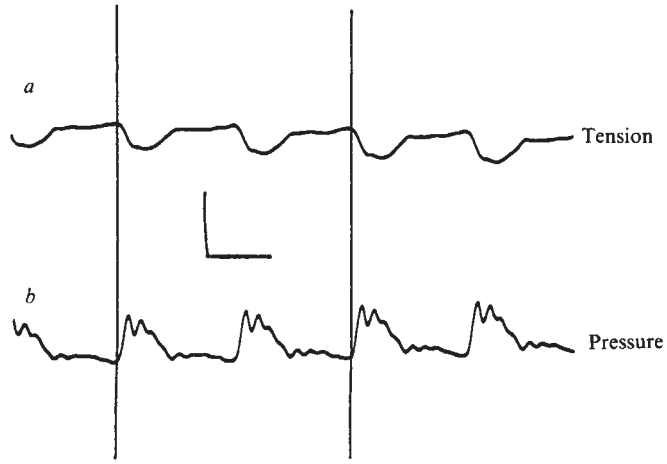

Fig. 2 Simultaneous recording of tension (*a*) and pressure (*b*) from the *in vivo* rabbit aorta. Time mark lines are drawn to coincide with the onset of the pressure wave. Scale bar: 0.6 g, 30 mm, 120 ms; basal blood pressure is 76.0 mm. An upward deflection reflects an increase in tension and pressure.

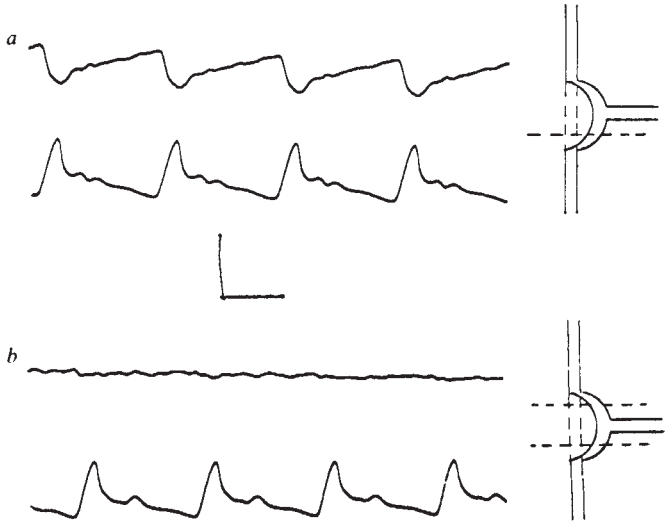

Fig. 3 Simultaneous recording of tension (upper) and pressure (lower) from the *in vivo* rabbit aorta. In (*a*), the distal side of the bypassed segment was severed from the intact aorta; in (*b*), the proximal side was then cut. Scale bar: 0.3 g, 40 mm, 150 ms; basal blood pressure in both (*a*) and (*b*) is 85 mm.

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same velocity as the pulse pressure waves. The resulting synchrony between the pulse and contraction waves ensures that the aorta relaxes as pulse pressure increases and contracts during pulse pressure diastole.

The physiological function of the aortic oscillations remains a topic for conjecture. A previous study[17] has suggested that the aortic arch may undergo a contraction–relaxation cycle with a 1:1 correspondence with the heartbeat, possibly aiding in the diastolic propulsion of blood. Another role may be to reduce mechanical stress on the aortic wall. Prokop *et al.*[18] showed that the rate of change of blood pressure, but not its absolute magnitude, is the primary cause of aortic rupture. Thus, the observed relaxation during the increase in pulse pressure may locally reduce the rate of change of blood pressure and protect the aortic wall from mechanical damage.

This research was supported by American Heart Association grant 781124, NIH grants HL 07188 and HL 23932, and American Heart Fellowship RF 1. We thank Barbara Bradie for typing the manuscript and Gina del Valle for technical assistance.

Received 8 September; accepted 31 December 1980.

1. Bazett, H. C. *Am. J. Physiol.* **70**, 550–585 (1924).
2. Maltby, A. B. & Wigers, C. J. *Am. J. Physiol.* **100**, 604–613 (1932).
3. Hamilton, W. F. & Dow, P. *Am. J. Physiol.* **125**, 48–59 (1939).
4. Alexander, R. S. *Fedn Proc.* **11**, 738–749 (1952).
5. Furchgott, R. F. & Bhadrakom, S. *J. Pharmac. exp. Theor.* **108**, 129–143 (1953).
6. Biamino, G. & Kruckenberg, P. *Am. J. Physiol.* **217**, 376–352 (1969).
7. Altura, B. M. & Altura, B. T. *Microvascular Res.* **7**, 145–155 (1974).
8. Furchgott, R. F., Ehrreich, S. J. & Greenblatt, E. *J. gen. Physiol.* **44**, 499–519 (1961).
9. van Breemen, C. *et al. Phil. Trans. R. Soc.* **B265**, 57–71 (1973).
10. Deth, R. & van Breemen, C. *J. Membrane Biol.* **40**, 363–380 (1977).
11. Fischer, E. *Physiol. Rev.* **24**, 467–490 (1944).
12. Dow, P. & Hamilton, W. F. *Am. J. Physiol.* **125**, 60–65 (1939).
13. Mountcastle, V. B. (ed.) *Medical Physiology* Vol. 1, 13th edn (Mosby, St Louis, 1974).
14. Ganong, W. F. *Review of Medical Physiology* 2nd edn (Lange Medical, Los Altos, 1969).
15. Tomita, T. *Prog. Biophys. molec. Biol.* **30**, 185–202 (1975).
16. Connor, J. A., Mangel, A. M. & Nelson, B. *Am. J. Physiol.* **237**, C237–C246 (1979).
17. Boucek, R. J., Takashita, R. & Fojaco, R. *Am. J. Anat.* **114**, 273–282 (1964).
18. Prokop, E. K., Palmer, R. F. & Wheat, M. W. *Circulation Res.* **27**, 121–127 (1970).

No need for a new membrane model

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Schindler *et al.*[1] recently reported lateral diffusion measurements in reconstituted membranes of phospholipid (PL), lipopolysaccharide (LPS) and *Escherichia coli* matrix protein (P), using the technique of fluorescence recovery after photobleaching (FRAP). Evaluation of their data led Schindler *et al.* to conclude that the fluid mosaic model is an inadequate description of the membrane and to propose a new membrane model. Their conclusion was based on identifying the fluid mosaic model with a particular binding behaviour of LPS to matrix protein. I present here a more general model for the association of membrane components, and demonstrate the use of lateral diffusion data in elucidating membrane structure. The data of Schindler *et al.* are shown to be reasonably interpretable on the basis of an association of LPS and matrix protein, which obviates the necessity for postulating a new membrane model.

I begin with a discussion of the lateral diffusion in a mixed membrane observed by the FRAP technique. Schindler *et al.*[1] studied the lateral diffusion constant D_S of one component such as LPS or PL, termed substrate, at different protein concentrations. The substrate molecules may be subdivided into those which are bound to protein and do not exchange in the time scale of a FRAP measurement, and the rest. D_S is given by the contributions from the non-exchanging molecules of molar fraction n_{nonex} and the remainder of the molar fraction n_{ex}: $D_S = D_P\, n_{nonex} + D_{ex}\, n_{ex}$. The diffusion constant of the non-exchanging molecules equals the protein diffusion constant D_P, which is usually much smaller than the diffusion constant of LPS or PL. If D_P vanishes, the fraction n_{nonex} of the substrate molecules does not diffuse, and the fluorescence recovery r is <100% and given by $r = n_{ex}$. In the measurements of Schindler *et al.*[1] the PL recovery was 100% and the LPS recovery >75%, implying that some LPS molecules were fixed to matrix protein and that the matrix protein diffusion was negligibly small. D_P was indeed found to be $<10^{-12}\ \mathrm{cm^2\,s^{-1}}$. In my treatment I will assume for simplicity that non-exchanging molecules do not exist: $n_{nonex} = 0$ and $n_{ex} = 1$, so that $D_S = D_{ex}$.

The molecules denoted as exchanging may diffuse completely freely or may interact with proteins. In the latter case, they may bind to proteins and thus become immobilized for some time, but then exchange with free molecules. Assuming the exchange to be rapid on the time scale of a FRAP experiment ($t > 1$ s), the diffusion constant D_{ex}, which is the temporal average over the free and bound states of a molecule, equals an ensemble average over the exchanging molecules at any time: $D_{ex} = D_f n_f + D_P n_b$. Here D_f is the diffusion constant for free diffusion, and the diffusion constant of the bound molecules is again equal to the protein diffusion constant D_P; n_f and n_b are the molar fractions of free and bound molecules with $n_f + n_b = 1$. I assume $D_P = 0$. Furthermore, D_f is regarded as independent of protein concentration, which is valid only if proteins do not affect the viscosity of the free-substrate phase. I introduce a normalized diffusion constant $\tilde{D}_S(\mathrm{P}) = D_S(\mathrm{P})/D_S(0)$, which is then simply given by the concentration of free substrate:

$$\tilde{D}_S(\mathrm{P}) = n_f(\mathrm{P}) \tag{1}$$

The function $n_f(\mathrm{P})$, a static quantity reflecting association of substrate and protein, can be derived from the law of mass action. We assume a simple binding equilibrium $\mathrm{S} + \mathrm{P}' \rightleftarrows \mathrm{SP}'$, where P' denotes protein-binding sites. The dissociation constant K_d is thus

$$K_d = \frac{\mathrm{S_f P'_f}}{\mathrm{S_b}} \tag{2}$$

The concentration of free binding sites is given by $\mathrm{P'_f} = \mathrm{P'_{tot}} - \mathrm{S_b}$, where $\mathrm{P'_{tot}}$ is the total concentration of binding sites. The latter equals $b\mathrm{P}$, with b representing the number of binding sites per protein molecule. Equation (2) can be transformed into

$$\mathrm{S_b} = b\mathrm{P}\,\frac{\mathrm{S_f}}{K_d + \mathrm{S_f}} \tag{3}$$

Insertion of $\mathrm{S_b} = \mathrm{S} - \mathrm{S_f}$, where S is the total substrate concentration, produces a quadratic equation for $\mathrm{S_f}$ which can be solved to yield the following expression for $n_f = \mathrm{S_f/S}$:

$$n_f = \tfrac{1}{2}(1 - b\mathrm{P/S} - K_d/\mathrm{S})\left(1 \pm \left[1 + \frac{4K_d/\mathrm{S}}{(1 - b\mathrm{P/S} - K_d/\mathrm{S})^2}\right]^{1/2}\right) \tag{4}$$

The sign has to be chosen so that $n_f \geq 0$. Note that n_f depends on the protein concentration via the molar ratio P/S, which will therefore be used as the measure of protein concentration. Equations (1) and (4) represent the variation of the lateral diffusion of LPS or PL with protein concentration.

To resolve further this result we discuss two limiting cases: first, the case of small protein concentration $b\mathrm{P/S} \ll 1$. Here equation (4) becomes

$$n_f = 1 - \frac{b\mathrm{P}}{\mathrm{S}}\,\frac{\mathrm{S}}{K_d + \mathrm{S}} \tag{5}$$

which is a functional relationship well known from enzyme kinetics. It describes the saturation of a few protein-binding sites with substrate where the number of bound molecules is proportional to the protein concentration. This implies that the substrate diffusion constant $\tilde{D}_S$ decreases linearly with protein

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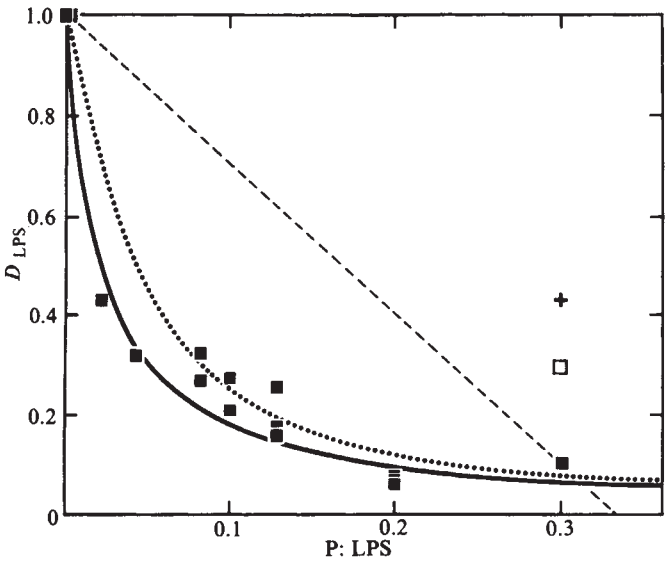

Fig. 1 Normalized lateral diffusion coefficient $\tilde{D}_{LPS}$ of LPS in membranes of PL, LPS and matrix protein as a function of the molar ratio P/LPS. Experimental data of Schindler *et al.*[1] for molar ratios PL/LPS of 6 (■) and 72 (□); for the conversion of weight % protein and weight ratio PL/LPS (given by Schindler *et al.*[1]) into molar ratios, the molecular weights $MW_P = 36{,}000$, $MW_{LPS} = 3{,}600$ and $MW_{PL} = 600$ were used. Theoretical curves are shown for PL/LPS = 6 in the limit of high P, equation (6), with $\kappa b = 325$ (—); in the general case, equation (4), with $\kappa b = 325$ and $b = 21$ (···); and in the limit of low P, equation (5), with $K_d = 0$ and $b = 3$ (---); and a theoretical point for PL/LPS = 72 in the limit of high P with $\kappa b = 325$(+).

concentration at constant substrate concentration. The second limiting case is that of high protein concentration $bP/S \gg 1$, where equation (4) yields

$$n_f = \frac{K_d}{K_d + bP} \tag{6}$$

This describes the situation where an excess of protein binds the few substrate molecules, but only succeeds in binding all of them asymptotically at infinite protein concentration. In this case there is saturation of the few substrate molecules by protein, which is the opposite of the first case. Here the substrate diffusion constant $\tilde{D}_S$ shows an asymptotic decrease to zero with increasing protein concentration.

Thus in this simple binding model there is a linear decrease in the substrate diffusion constant at low protein concentration, due to protein saturation, and a gradual decrease to zero at high protein concentration, due to substrate saturation. Experimental results for $\tilde{D}_S$ can be evaluated to yield the number b of protein-binding sites and the dissociation constant K_d.

The variation of $\tilde{D}_S$ with protein and substrate concentration derived above can also be formulated in another model which includes a partition coefficient. Two possible advantages of such a model are: first, it may be more appropriate in the case of many protein binding sites, $b \gg 1$; second, it may lend itself to further generalization concerning interactions between substrate and membrane components other than protein. Consider the partitioning of a substrate molecule between protein and lipid consisting of PL and LPS: the probability p_P that a substrate molecule is bound to protein is proportional to the concentration of free protein-binding sites (P′), and the probability p_L of association with lipid is proportional to the concentration of free lipid sites (L), so that

$$\frac{p_P}{p_L} = \frac{P'_{tot} - P'_b}{L_{tot} - L_b} \cdot \kappa \tag{7}$$

where κ is the partition coefficient. The substrate molecules in the lipid phase have been denoted free, therefore $p_P/p_L = S_b/S_f$ at equilibrium. On the right-hand side of equation (7) we can assume $L_{tot} \gg L_b$, and substitute bP for P'_{tot} and S_b for P'_b. Solving for S_b we obtain a relation which is identical to equation (3) and has the same solution equation (4), with the substitution

$$K_d = L_{tot}/\kappa \tag{8}$$

Thus the partition coefficient model leads back to the binding model. However, due to the appearance of the lipid-binding sites, L_{tot}, the partition coefficient model can also account explictly for the interaction of substrate, for example LPS, with PL and LPS. The simplest approximation is

$$L_{tot} = PL + LPS \tag{9}$$

The final result for $\tilde{D}_S$ is obtained in the form of equation (4) for n_f with the substitution of K_d according to equations (8) and (9). In the limiting case of high protein concentration as in equation (6), the following is obtained:

$$\tilde{D}_S\left(\frac{P}{LPS}, \frac{PL}{LPS}\right) = 1 \Big/ \left(1 + \frac{\kappa b}{1 + PL/LPS} \frac{P}{LPS}\right) \tag{10}$$

These theoretical results can now be used to evaluate the experimental data of Schindler *et al.*[1] for the lateral diffusion of LPS. They are presented as $\tilde{D}_{LPS}$ (P/LPS) in Fig. 1. The data points for PL/LPS = 6 show a gradual decrease to zero with increasing protein concentration indicating the limiting case of high protein concentration. To test this suggestion we plotted $\tilde{D}^{-1}_{LPS}$ against P/LPS (see Fig. 2) which should yield a straight line according to equation (10). Although the data points scatter appreciably, they can be regarded as conforming to a straight line; a least-squares fit gives $\kappa b = 325$. There is no indication of a cross-over of the experimental points from the limit of high protein concentration to the case of low protein concentration, equation (5); therefore the quantitites b and κ (or K_d) cannot be determined separately. A lower limit for b may be estimated from the condition of high protein concentration, $bP/LPS \gg 1$. Assuming that this condition conforms to the experimental point at P/LPS = 0.047 in Fig. 2, we obtain $b \gg 21$. The theoretical curve for $\tilde{D}_{LPS}$ with $b = 21$, derived from the general

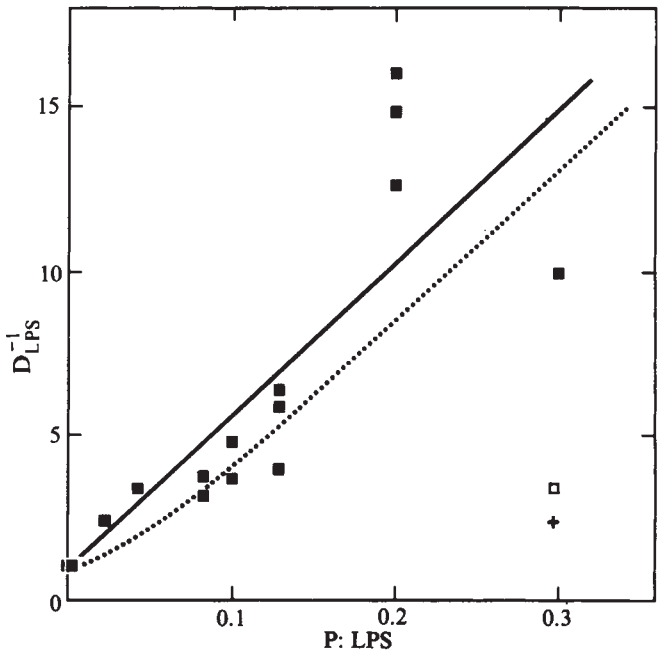

Fig. 2 Inverse of the normalized lateral diffusion coefficient $\tilde{D}_{LPS}$. Representation of points and lines as in Fig. 1.

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expression (4), is included in Figs 1 and 2. Comparison with the earlier fit based on equation (10), which may be regarded as the limiting case for large b, shows that $b = 21$ represents a lower limit for b. Such a high number of binding sites does not seem unlikely because such binding is only a loose association between LPS and protein, comparable with lipid–protein association, where even higher numbers of boundary lipids associated with protein are known.

In contrast to the evaluation presented here, Schindler *et al.*[1] restricted themselves to $b = 3$ and $K_d = 0$. With these values equation (6) yields $\tilde{D}_{LPS} = n_f = 0$ at high protein concentration, and at low protein concentration equation (5) yields a straight line for $\tilde{D}_{LPS}$ (P/LPS). The latter is included in Fig. 1, showing its serious disagreement with the experimental data, which led Schindler *et al.*[1] to propose a new membrane model. They adopted the number of three binding sites from studies of the *E. coli* outer membrane, where it was found that three LPS molecules are associated with one matrix protein[2]. However, the assumption $b = 3$ would imply that the matrix proteins are saturated with LPS at the relatively high protein concentration of the outer membrane, which seems rather unlikely. Within our evaluation we would predict from equation (10), with $\kappa b = 325$ and the outer membrane molar ratios P/LPS = 1/3 and PL/LPS = 6, that $n_f = 0.06$, or 2.82 LPS molecules are associated with 1 matrix protein. This number for the site occupation agrees well with the experimental result, while the number of binding sites is expected to be much higher, as discussed. Note, however, that the evaluation of Schindler *et al.* and the one presented here differ not only in the number of binding sites, but also in the binding constant, producing a different dependence of $\tilde{D}_{LPS}$ on protein concentration.

Because the evaluation of Schindler *et al.* is too restricted, their conclusions concerning old and new membrane models are invalid. However, I hope I have made it clear that lateral diffusion measurements such as those of Schindler *et al.* are useful in studying the association behaviour of membrane constituents.

I thank many of my colleagues for stimulating discussions.

Received 30 June; accepted 1 December 1980.

1. Schindler, M., Osborn, M. J. & Koppel, D. E. *Nature* **283**, 346–350 (1980).
2. Schindler, H. & Rosenbusch, J. P. *Proc. natn. Acad. Sci. U.S.A.* **75**, 3751–3755 (1978).

Reply from D. E. Koppel*, M. J. Osborn† & M. Schindler‡

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Some comments are in order to put Dr Jähnig's re-evaluation of our data in a proper perspective. We determined the lateral diffusion coefficients of lipopolysaccharide (LPS), phospholipid (PL) and *Escherichia coli* matrix porin protein in reconstituted multibilayer membranes[1], using the technique of fluorescence redistribution after photobleaching. We compared the results from a series of samples of constant relative LPS and PL content (1:1 by weight), but different protein content (0–60% by weight). It was found that as the protein concentration increased, the diffusion coefficient of LPS decreased by an order of magnitude, whereas that of PL decreased to a much smaller extent. The protein molecules at the higher concentrations were essentially 'immobile', with little or no redistribution observable over the distance scale (~3 μm) and maximum time scale (~3×10^3 s) of these experiments. This immobility would be a surprising result in the context of the fluid mosaic model, but is not unexpected in view of considerable evidence demonstrating the self-association of porin trimers into ordered aggregates *in vivo* as well as *in vitro*[2].

Jähnig has chosen to interpret these results solely in terms of specific LPS binding to the immobile protein, leaving both the dissociation constant and the stoichiometry of binding as floating parameters. This analysis can yield results consistent with the data, but only if one allows a total of at least 21 LPS binding sites per protein monomer (that is, ~63 sites per trimer).

In contrast to the above, we[1] did not assume that in the absence of specific binding the diffusion rates would be necessarily unaffected by the presence of an immobile protein matrix in the membrane, but set out to try to determine what other factor, if any, could be contributing to the observed diffusion characteristics. We evaluated the possible effect of LPS binding consistent with that which we considered to be the best available value of binding stoichiometry. Based on evidence of co-purification, we estimate this as 9 LPS molecules per protein trimer[3,4]. Under this constraint, the binding model of diffusion could not account for the magnitude of the observed decrease in LPS diffusion. As a possible explanation of this difference we speculated that the translational diffusion of membrane components in these conditions might bear a closer resemblance to diffusion through a polymeric network than to diffusion in a simple viscous fluid. The differential effects observed for LPS and PL would be explained by the different sizes of the diffusing species. The part of the polymeric network in this scenario is played by the immobile protein matrix. Subsequent experiments[5] have suggested that the peripheral protein matrix of erythrocyte membranes may exert a similar control over the diffusion of integral membrane proteins projecting beyond the membrane bilayer.

Evidence is not yet available to allow a final definitive analysis. The value of the stoichiometry we have selected is, admittedly, a relatively soft number—it could be higher. At the same time, the value deduced by Jähnig seems unreasonably high for a specific binding interaction. The truth probably lies somewhere in between.

1. Schindler, M., Osborn, M. J. & Koppel, D. E. *Nature* **283**, 346–350 (1980).
2. Osborn, M. J. & Wu, H. C. P. *A. Rev. Microbiol.* **34**, 369–422 (1980).
3. Schindler, H. & Rosenbusch, J. P. *Proc. natn. Acad. Sci. U.S.A.* **75**, 3751–3755 (1978).
4. Rosenbusch, J. P., Steven, A., Alkan, M. & Regenass, M. in *Regular 2-D Arrays of Biomolecules: Structure Determination and Assembly* (ed. Baumeister, W.) (Springer, Berlin, 1979).
5. Sheetz, M. P., Schindler, M. & Koppel, D. E. *Nature* **285**, 510–512 (1980).

Molecular cloning of the K1 capsular polysaccharide genes of *E. coli*

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Epidemiological and immunological evidence indicates that the K1 capsular polysaccharide confers the property of virulence on *Escherichia coli*. *E. coli* K1 is associated with invasive diseases in humans and in laboratory and domesticated animals[1]. K1 isolates account for 80% of *E. coli* neonatal meningitis and comprise the majority of capsular types in neonatal septicaemia without meningitis and in childhood pyelonephritis[2,3]. Passive administration of K1 antibodies prevented bacteraemia and meningitis in infant rats fed *E. coli* K1[4]. Nonencapsulated

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derivatives of these invasive K1 strains did not cause bacteraemia in infant rats, although intestinal colonization was similar to that of the parent strains (M. Achtman and R.P.S., unpublished results). Several reports propose that the *E. coli* K1 capsular polysaccharide exerts an anti-phagocytic effect similar to that observed with other pathogenic encapsulated bacteria[5,6]. One approach to studying whether the K1 antigen is sufficient to confer virulence or if other *E. coli* structures are necessary is to isolate the K1 genes for genetic and biochemical analysis. Recombinant DNA methodology provides a powerful tool for such an approach. Here, we report the molecular cloning of the *E. coli* K1 antigen genes. The cloned K1 genes synthesize a capsule in *E. coli* K12 indistinguishable chemically and immunologically from that of wild-type K1 strains.

The K1 capsular polysaccharide is a linear homopolymer of α2–8-linked *N*-acetylneuraminic acid (NANA)[7,8]. A genetic locus, associated with K1 biosynthesis, linked to the *serA* locus on the *E. coli* chromosome and termed *kpsA*, has been described elsewhere[9]. A membrane-associated sialyltransferase complex catalyses the incorporation of sialic acid from CMP-sialic acid into polymeric products[10]. The number and function of the genes involved and the molecular events controlling the synthesis and assembly of the K1 polysaccharide have not been elucidated. Therefore, we chose to use the cosmid cloning vector pHC79 in conjunction with *in vitro* λ packaging techniques to clone the K1 genes[11,12]. The plasmid pHC79, a 6.4-kilobase derivative of pBR322, confers resistance to ampicillin (Amp) and tetracycline (Tc)[11]. *E. coli* RS218 (ref. 13) (*O*18, K1, H7) chromosomal DNA[14] (75 μg ml^{-1}), partially digested with *Bam*HI, and *Bam*HI-cleaved pHC79 (225 μg ml^{-1}), were mixed and ligated in a total volume of 20 μl (ref. 12). *Bam*HI inactivates the Tc resistance gene of pHC79. Ligated DNA (8 μl) was packaged in 400 μl of packaging lysate[12]. *E. coli* K12 strain HB101 (ref. 15) (*hsd*R, *hsd*M, RecA, *leu pro*) was transduced by diluting the packaging mix to 2.0 ml with L-broth[16](LB) (containing 200 mM $MgSO_4$) and adding the mixture to 10 ml of cells (10^9 per ml in LB grown in 0.4% maltose). After 10 min adsorption at 30 °C the mixture was diluted five-fold with LB, incubated for 1 h at 37 °C, concentrated 10-fold and plated on L-agar[16] or antiserum agar[17] containing 50 μg ml^{-1} ampicillin. Resistance to Tc was determined by growth on L-agar containing 20 μg ml^{-1} Tc. Complementation of proline auxotrophy was detected by the ability of HB101-containing hybrid plasmids to grow in minimal medium supplemented with leucine but lacking proline.

Amp-resistant transductants (4×10^4 per μg ligated *E. coli* DNA) were obtained from the L-agar plates. Thirty per cent of the transductants were Tc sensitive and 1.4% were proline prototrophs, confirming the insertion and expression of *E. coli* chromosomal DNA into the *Bam*HI site of pHC79. Amp^R colonies (6×10^3) were pooled from L-agar plates and stored at −20 °C. These cells were later grown in LB and their plasmid DNA isolated. Contour length measurements[18] of purified plasmid DNA showed a homogeneous population of 43.8 kilobases (14.78 ± 0.57 μm). This is consistent with sizes previously reported for cosmid hybrids[11].

The K1 antigen in the transduced cells was detected by halo (precipitin) formation on antiserum agar plates containing equine meningococcal group B antiserum (the K1 capsular polysaccharide is structurally and antigenically identical to the meningococcal group B polysaccharide)[8]. There were fewer Amp^R transductants per μg ligated *E. coli* DNA on antiserum agar than on L-agar and no halo-producing colonies were observed among more than 1,500 transductants. In contrast, when Amp^R cells from the L-agar colony pool were plated on antiserum agar, many halo-producing colonies were isolated. Our inability to isolate transductants containing K1, except after plating on medium lacking antiserum, may reflect a delay in the expression of the newly acquired K1 genes. *E. coli* K12 was sensitive to the bactericidal effects of the antiserum; Table 1 shows a 10^5-fold decrease in plating efficiency of HB101 on antiserum agar. The bactericidal effect was eliminated by heating the serum at 56 °C for 30 min, suggesting that serum sensitivity was due to complement proteins. *E. coli* K12 expressing the K1 antigen was not serum sensitive (Table 1). This is probably due to shielding effects of the capsule on deeper cell structures capable of activating complement proteins.

Another factor contributing to the difficulty in isolating K1 transductants was the instability of pHC79/K1 hybrid plasmids. We observed a 10^4-fold decrease in the number of K1$^+$Amp^R organisms following overnight growth in the absence of antibiotic selection. The instability of the plasmid is consistent with previous observations. Henning *et al.*[19] failed to find the *OmpA* gene in the *E. coli* hybrid gene bank of Clark and Carbon[20]. Nishimira *et al.*[21] also failed to find complementation of other outer membrane protein mutants in the Clark and Carbon[20] collection including the *OmpC* and lipoprotein genes. Henning *et al.*[19] were unable to clone the *OmpA* gene on a multicopy vector; *OmpA* was cloned on λ and pSC101 vectors. These data suggest that the gene dosage of outer membrane proteins and other surface structures, generated by cloning on multicopy

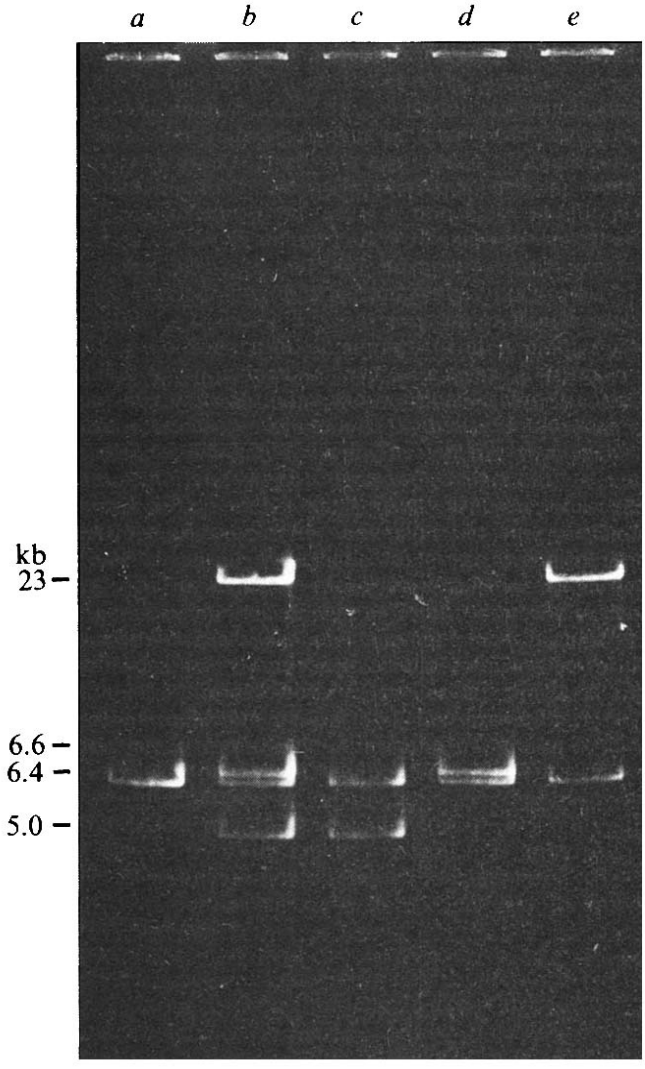

Fig. 1 Electrophoresis in 0.7% agarose gel of *Bam*HI-digested plasmids. Plasmid DNA was isolated as described elsewhere[13] and treated with endonuclease *Bam*HI in 100 mM Tris-HCl (*p*H 7.5), 10 mM $MgCl_2$ and 50 mM NaCl at 37 °C for 1 h. Samples were run in 0.7% agarose gels, Tris-borate buffer, *p*H 8.2, for 3 h at 120 V. DNA fragments were visualized by staining with ethidium bromide (0.4 μg ml^{-1}) and photographed in short-wavelength UV light. Molecular size standards of *Eco*RI-cleaved λ DNA were used to indicate fragment length. Lane *a*, pHC79; lane *b*, pSR23; lane *c*, pSR25; lane *d*, pSR26; lane *e*, pSR27. pSR25–27 were isolated following *Bam*HI digestion of pSR23, re-ligation[12] and transformation[22] of HB101. kb, Kilobases.

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plasmid vectors, may be directly related to the lethality of the cell[19]. A recent observation that a strain in which the K1 genes have been transposed to a low copy number plasmid (F*lac*Mu) stably produced the K1 antigen is consistent with this view (R.P.S., unpublished observation).

The production of the K1 antigen could be maintained in broth cultures or on agar plates containing 500 or 100 $\mu g\ ml^{-1}$ ampicillin, respectively. A 40-kilobase ($13.7 \pm 0.48\ \mu m$) plasmid, designated pSR23, was isolated from HB101K1$^+$ cells. A *Bam*HI digest of pSR23 generated four fragments: the 6.4 vector and three *E. coli* fragments of 23, 6.6 and 5.0 kilobases (Fig. 1). A DNA–DNA heteroduplex between pHC79 and pSR23 confirmed the presence of a 35-kilobase insert in pHC79 (Fig. 2). HB101 was readily transformed[22] to ampicillin resistance by pSR23; all Amp^R transformants produced the K1 antigen. The efficiency of plating of K1 specific phage[23] was the same on HB101(pSR23) as on RS218. The 35-kilobase cloned *E. coli* fragment represents about 1 min of the *E. coli* genetic map[24]. It contains information to code for about 50 proteins and is larger than that required for K1 biosynthesis alone. Other *E. coli* genes on the hybrid plasmid have not been identified. Subclones containing pHC79 and each of the *Bam*HI fragments of pSR23 (Fig. 1, lanes *c–e*) did not produce K1 polysaccharide as determined by halo formation on antiserum agar and K1 phage sensitivity. Cells harbouring pSR27, which contains the largest *Bam*HI fragment (Fig. 1, lane *e*), synthesized NANA and had sialyltransferase activity (W.F.V. and R.P.S., unpublished results).

The structure of the K1 polysaccharide isolated from HB101(pSR23) was studied by several techniques[25]. We found that: (1) it was composed of 88% sialic acid; (2) the molecular size, determined by gel filtration, was similar ($k_d = 0.5$) to that of K1 polysaccharides isolated from other K1 strains; (3) the purified polysaccharide was negatively charged at neutral *p*H; (4) in double immunodiffusion a line of identity was observed between the polysaccharide from HB101(pSR23) and wild-type K1; and (5) the ^{13}C-NMR spectrum of the HB101(pSR23) polysaccharide was identical to that of wild-type K1.

Isolation of the K1 genes will permit a direct examination of whether the K1 capsular polysaccharide alone confers the property of invasiveness to *E. coli*. Our preliminary experiments indicate that *E. coli* K12 strains with the K1 capsular polysaccharide genes are not invasive for the infant rat. Thus, it is likely that biochemical traits in addition to the K1 capsule are required for the invasiveness of *E. coli*. The techniques of molecular biology and bacterial genetics should provide useful approaches to characterizing these pathogenic traits.

We thank John B. Robbins and M. Carolyn Hardegree for reviewing the manuscript, William Egan for NMR studies, John Collins for providing pHC79, B. Rowe for providing K1 specific phage, and Helen C. Coon for assistance in preparation of the packaging extract.

Table 1 Growth of *Escherichia coli* on antiserum agar

Strain	Plating efficiency
RS218	1.0
HB101	10^{-5}
HB101 (serum heated 56 °C, 30 min)	1.0
CSH75*	10^{-4}–10^{-5}
CSH75K1$^+$†	0.5

Cells were grown overnight in LB at 37 °C, diluted and plated on antiserum agar and L-agar. Efficiency of plating on antiserum agar was expressed relative to growth on L-agar.

* *E. coli* K12 strain (*ara, leu, lacY, purE gal trp his, argG malA, strA, xyl, mtl, ilv, met, thi, pro*).

† Chromosomal recombinant obtained from a mating with an *E. coli* K1 Hfr donor (R.P.S. and J. Foulds, unpublished).

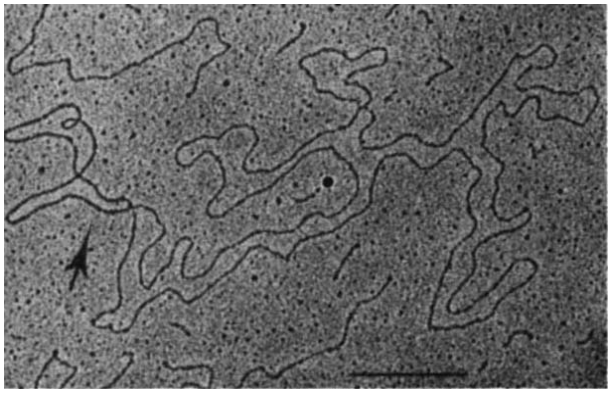

Fig. 2 Electron micrograph of DNA–DNA heteroduplex between pHC79 and pSR23. pHC79 was digested with endonuclease *Pst*I at 37 °C in 20 mM Tris-HCl (*p*H 7.5), 10 mM $MgCl_2$, 50 mM $(NH_4)_2SO_4$ generating linear molecules. pSR23 was incubated with pancreatic DNase I (0.02 units ml^{-1}) for 25 min at 25 °C, conditions producing approximately one nick per molecule. Heteroduplex analysis was performed as described previously[18]. Arrow indicates double-stranded 6.4-kilobase pHC79. Scale bar, 1 μm.

Received 29 August; accepted 3 December 1980.

1. Schiffer, M. S. *et al. Pediat. Res.* **10,** 82–87 (1976).
2. Robbins, J. B. *et al. New Engl. J. Med.* **290,** 1216–1220 (1974).
3. Kaijser, B., Hanson, L. A., Jodal, V., Linden-Janson, G. & Robbins, J. B. *Lancet* **i,** 663–664 (1977).
4. Glode, M., Sutton, A., Moxon, R. & Robbins, J. B. *Infect. Immun.* **16,** 75–80 (1977).
5. Bortolussi, R., Ferrieri, P., Bjorksten, B. & Quie, P. G. *Infect. Immun.* **25,** 293–298 (1979).
6. Weinstein, R. & Young, L. S. *J. clin. Microbiol.* **8,** 248–255 (1978).
7. Barry, G. T. *J. exp. Med.* **107,** 507–521 (1959).
8. Kasper, D. L., Winkelhake, J. L., Zollinger, W. D., Brandt, B. L. & Artenstein, M. S. *J. Immun.* **110,** 262–268 (1973).
9. Orskov, I., Sharma, V. & Orskov, F. *Acta path. microbiol. scand.* **B84,** 125–131 (1976).
10. Troy, F. A. *A. Rev. Microbiol.* **33,** 519–560 (1979).
11. Collins, J. *Meth. Enzym.* **68,** 309–326 (1979).
12. Kretschmer, P. J. *et al. J. biol. Chem.* **255,** 3204–3211 (1980).
13. Silver, R. P., Aaronson, W., Sutton, A. & Schneerson, R. *Infect. Immun.* **29,** 200–206 (1980).
14. Smith, M. G. *Meth. Enzym.* **12,** 545–550 (1967).
15. Boyer, H. W. & Roulland-Dussoix, D. *J. molec. Biol.* **41,** 459–472 (1969).
16. Miller, J. *Experiments in Molecular Genetics* (Cold Spring Harbor Laboratory, New York, 1972).
17. Sarff, L. D. *et al. Lancet* **i,** 1099–1104 (1975).
18. Davis, R. W., Simon, M. & Davidson, N. *Meth. Enzym.* **21,** 413–428 (1971).
19. Henning, V., Royer, H-D., Teather, R. M., Hindennach, I. & Hollenberg, C. *Proc. natn. Acad. Sci. U.S.A.* **76,** 4360–4364 (1979).
20. Clarke, L. & Carbon, J. *Cell* **9,** 91–99 (1976).
21. Nishimura, Y. *et al. Plasmid* **1,** 67–74 (1977).
22. Cohen, S. N., Chang, A. C. Y. & Hsu, L. *Proc. natn. Acad. Sci. U.S.A.* **69,** 2110–2114 (1972).
23. Gross, R. J., Cheasty, T. & Rowe, E. *J. clin. Microbiol.* **6,** 548–550 (1977).
24. Bachmann, B. J. & Low, K. B. *Microbiol. Rev.* **44,** 1–56 (1980).
25. Ørskov, F. *et al. J. exp. Med.* **149,** 669–685 (1979).

Erratum

In the letter 'Relative departures in July temperatures in northern Canada for the past 6,000 yr' by J. T. Andrews *et al.*, *Nature* **289,** 164–167 (1981), Figs 2 and 3 have been incorrectly numbered. Figure 2 is the graph of relative departures of average July temperatures from their means and Fig. 3 a time-series of estimated July temperatures.

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MATTERS ARISING

Brown adipose tissue and diet-induced thermogenesis

IT has been proposed[1,2] that 'diet-induced thermogenesis' in brown adipose tissue enables animals to dissipate excessive intake of energy and so avoid obesity. The hypothesis is claimed to justify the view that thermogenesis is more important in regulation of energy balance than control of food intake[3], and the report by Rothwell and Stock[1] has been presented by the media (a BBC Television programme *Horizon*, 10 December 1979) as embodying an important discovery in relation to human obesity. We consider their data and arguments inadequate to support the hypothesis for the following reasons:

(1) The first paper hinges on an experiment in which one group of six rats was induced to overfeed by 'cafeteria feeding' (that is, providing a variety of tasty foods)[4,5] for 3 weeks, with a second group as controls. The amount of energy the cafeteria-fed rats stored as fat was much less than the increase in their intake of energy—this discrepancy supposedly demonstrated thermogenesis. We consider that it is hazardous to infer energy expenditure in this way (see Table 1 of ref. 1), particularly where it is the quantity of primary interest. (The paper includes in Table 2 some sample measurements of oxygen consumption as 'confirmation of lowered efficiency', but energy expenditure is too variable for short measurements to be useful in balance experiments, and the adjustment of the data for body weight complicates interpretation.) Accurate measurements of energy intake, expenditure and storage over a balance period are laborious and technically exacting; any systematic errors, however, cumulate in balance experiments, thus unless all three components of energy balance are measured there is no check on accuracy.

(2) Metabolizable energy intake (that is, the energy in food supplied minus the losses due to scattering and in excreta) cannot be reliably predicted from food composition tables or prior measurements: bomb calorimetry of the actual materials is essential. This must apply particularly to cafeteria feeding, where a variety of proprietary foods of high energy density are offered.

(3) Energy storage was determined by subtracting initial from final carcass energy values, determined by *post mortem* analysis; the method and its precision are not stated. The initial values were obtained from a group of six rats, killed at the start; their body composition was stated to be identical with that of the two groups used in the experiment, on the basis of *in vivo* tritium dilution measurements. We believe that estimation of body fat by *in vivo* body water measurement is inevitably inaccurate, and that this view is confirmed by Rothwell and Stock's data[6] on the tritium method.

(4) Rothwell and Stock[7-9] previously reported that cafeteria-fed rats showed similar resting oxygen consumption to controls and rapid increase of weight; that is, there was no evidence of thermogenesis (though it was reported to occur briefly after cafeteria feeding had ended). There are two possible explanations, other than experimental error, for these contrasting results. Rothwell and Stock[1,10] state that rats obtained from two suppliers, although of the same strain, differed in their capacity for thermogenesis and propensity to obesity. The only difference quoted in absolute terms was that energy expenditures during cafeteria feeding were 390 and 340 kJ per day. This difference, though statistically significant, is not large, and might be within the margin of error. If it is real, the selection of rats from one source rather than the other as the basis for a general theory would seem to require justification.

(5) We believe the age of the rats may provide the explanation. In Rothwell and Stock's earlier work[7-9] female rats weighed ~300 g and males 400 g and controls gained weight at 0.5–1.0 g per day, values typical of adult rats. The rats used in the later experiment[1] were described as 'adult' but their age was not stated; the control group were gaining weight at 5 g per day which is a rate typical of young, actively growing rats. Elsewhere Rothwell and Stock[2,10] describe as 'adult' rats which were 6 weeks, 10 weeks and 75 days old. Growth is costly of energy and its rate is sensitive to the supply of energy; thus it is well known that young, actively growing animals do not acquire fat readily. We believe a discrepancy between increased energy intake and fat storage, seen in young rats but not in adults, is more likely to reflect the young animals' capacity to use energy for growth than to demonstrate a new mechanism for energy dissipation. If the young animals' capacity for growth were to be regarded as a regulatory mechanism for energy balance, it would have to be made clear that it is not one available to adults. In adults, deposition of fat, although a relatively efficient process, entails dissipation as heat of about a third as much energy as is stored[11-13]; any discrepancy found in adults between excess intake and storage would have to be shown to exceed this before any possibility of a new mechanism arose.

(6) Since the work of Dawkins, Hull and others in the 1960s[15,21] it has been accepted that brown fat, controlled by noradrenergic sympathetic nerves, is an important heat-producing organ in the regulation of body temperature in infant animals of many species. Existing evidence indicates that the amount of brown fat in the body, and the heat produced in response to noradrenaline, decline with age[14-17]. The suggestion that, in animals possessing it, brown fat might be activated in response to overfeeding is an interesting one, although in young animals such a response would be expected to be antagonistic to growth. Miller and Payne[18] and Gurr *et al.*[19] have reported that diet-induced thermogenesis is conspicuous in young pigs; the pig is thought to possess no brown fat[20]. In our view, however, better evidence is required that diet-induced thermogenesis exists at all, before the question of its mechanism is worth pursuing. Extension of the suggestion to humans, and to adults, (and so relevance to human obesity) would seem doubly doubtful. The pair of IR thermograms[1] presented are certainly not adequate evidence: the effect seen after administration of ephedrine could as readily be ascribed to circulatory changes (G. A. Brown, RAF Institute of Aviation Medicine, personal communication).

Rothwell and Stock's hypothesis may still prove correct. Our point is that a novel theory which reverses previously accepted views needs substantial supportive evidence that can stand up to critical evaluation. We do not think such evidence has yet been presented.

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1. Rothwell, N. J. & Stock, M. J. *Nature* **281,** 31–35 (1979).
2. Brooks, S. L., Rothwell, N. J., Stock, M. J., Goodbody, A. E. & Trayhurn, P. *Nature* **286,** 274–276 (1980).
3. Perkins, M. N., Rothwell, N. J., Stock, M. J. & Stone, T. W. *J. Physiol.* (in the press).
4. Sclafani, A. & Springer, D. *Physiol. Behav.* **17,** 461–471 (1976).
5. Rolls, B. J. & Rowe, E. A. *J. Physiol.* **272,** 2P (1977).
6. Rothwell, N. J. & Stock, M. J. *Br. J. Nutr.* **41,** 625–628 (1979).
7. Rothwell, N. J. & Stock, M. J. *J. Physiol.* **291,** 59P (1979).
8. Stock, M. J. & Rothwell, N. J. in *Animal Models of Obesity* (ed. Festing, M. F. W.) 141–151 (Macmillan, London, 1979).
9. Rothwell, N. J. & Stock, M. J. *J. comp. physiol. Psychol.* **93,** 1024–1034 (1979).
10. Rothwell, N. J. & Stock, M. J. *Proc. nutr. Soc.* **39,** 20A (1980).
11. Blaxter, K. L. in *Obesity in Perspective* (ed. Bray, G. A.) 127–135 (DHEW Pub. No. (NIH) 75–708, 1975).
12. Pullar, J. D. & Webster, A. J. F. *Br. J. Nutr.* **37,** 355–363 (1977).
13. van Es, A. J. H. *Nutr. Metab.* **21,** 88–104 (1977).
14. Alexander, G. *Int. Rev. Physiol.* **20,** 43–155 (1979).
15. Smith, R. E. & Horwitz, B. A. *Physiol. Rev.* **49,** 330–425 (1969).
16. Heaton, J. M. *J. Anat.* **112,** 35–39 (1972).
17. Himms-Hagen, J. *Pharmac. Rev.* **19,** 367–461 (1967).
18. Miller, D. S. & Payne, P. R. *J. Nutr.* **78,** 255–262 (1962).
19. Gurr, M. I., Mawson, R., Rothwell, N. J. & Stock, M. J. *J. Physiol.* **291,** 60P (1979).
20. LeBlanc, J. & Mount, L. E. *Nature* **217,** 77–78 (1968).
21. Dawes, G. S. *Foetal and Neonatal Physiology* Ch. 15 (Year Book, Chicago, 1968).

ROTHWELL AND STOCK REPLY—We shall deal with points (1), (2) and (3) above together. Estimation of energy expenditure by the carcass balance method (that is, intake minus storage) is an accepted

0028–0836/81/080699—02$01.00
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and reliable technique that has been in routine use in our laboratory for 15 years. We have introduced various improvements to increase accuracy and the coefficient of variation on carcass energy determination is less than 1% (ref. 1). A further improvement we have made is to match groups of rats at the start of the experiment (including those killed for determining initial carcass energy) on the basis of weight and body composition. In adult rats there is a poor correlation between body weight and energy content[2] and matching groups by weight alone or previous growth rate (as used by others, including Hervey and Tobin) can introduce very large errors. Obviously matching for body composition has to be done using *in vivo* measurements and, contrary to Hervey and Tobin's statement, our tritium method[3] gives values for body fat which do not differ from values determined by direct carcass analysis. However, after using the *in vivo* method for group matching, we then determine carcass composition of all groups (initial and final) by direct analysis, and this was clearly stated in our article[4].

We would share Hervey and Tobin's reservations about the use of food composition tables, but by carefully selecting appropriate foods and making meticulous measurements of food intake, metabolizable energy can be estimated conveniently and reliably. We have checked this against determinations by bomb calorimetry and the errors are small (always <5%)—this must be compared with the very large difference in energy intake (80%) on the cafeteria diet. All non-quantal measurements have errors and the skill of experimental design relies on producing differences that are not obscured by these errors. Thus, even if we take an extreme and unlikely 'worst case' example and assume the energy intake of cafeteria rats was overestimated by 10% and energy storage underestimated by 10%, we still find that cafeteria rats ate 62% more than controls. Of this additional intake, only 8% was stored and all the remainder was lost as heat—that is, as diet-induced thermogenesis (DIT).

Hervey and Tobin take us to task (point (4) above) for obtaining contrasting results in rats from different colonies, and yet they must be aware of the strain and intra-strain differences that are known to affect many aspects of metabolism (see for example, Miller[5]). Biologists cannot ignore these powerful genetic influences on metabolism, and selecting rats from different colonies is an attempt on our part to simulate in laboratory rats the wide individual variability seen in many animals, including man.

The rats used in our studies ranged from 200 to 400 g in weight (7–16 weeks of age). In terms of reproductive maturity these animals are past puberty and in anthropomorphic terms range from 'young adult' to almost 'middle-aged'. DIT can be induced at each age and to say (point (5) above) that age explains our results ignores the fact that rats of the same age can maintain weight on intakes that differ by up to 60% (ref. 6). If anything, our brown fat hypothesis might be used to explain why the incidence of obesity increases in later life when the amount and activity of brown fat is known to decline (refs 14–17 above). We cannot accept the argument that the energy cost of growth or fat deposition accounts for DIT for the following reasons. We have observed increased DIT when (i) the rate of growth is unaffected[6]; (ii) fat deposition is lower in one cafeteria group than another[7]; and (iii) when animals are hypophagic and losing body fat[4,8]. We have also calculated that the energy cost of increased fat deposition (when it occurs) accounts for less than 8% of the increased heat production[4] and even this assumes that all the fat was synthesized and none was derived from dietary sources.

We wonder why Hervey and Tobin accept the importance of brown fat in infant animals (point (6) above), for which there is no good quantitative evidence, and ignore the evidence provided by Foster and Frydman's meticulous study in cold-adapted adult rats[9]. Using their technique we provided similar quantitative evidence in cafeteria-fed rats[10] but they ignored this too, even though they are well aware of it. Our hypothesis is not intended to be exclusive and in our discussion of the pig experiments (ref. 20 above) we suggested alternative mechanisms of DIT in this animal which supposedly lacks brown fat, although it now appears that the young pig does possess some brown fat (D. L. Ingram, personal communication). The IR thermograms[4] are certainly not adequate to support a role for brown fat in adult man, but we doubt that 'circulatory changes' (personal communication, above) can explain how, after ephedrine, the temperature at the nape of the neck can rise above core temperature while other adjacent areas remain unaltered.

In terms of DIT, our work is not novel but merely confirmation of a phenomenon first observed at the turn of the century[11] and subsequently reported by many other workers (see ref. 4). Hervey and Tobin have ignored this earlier evidence and make no comment about the evidence for a thermogenic defect in genetic obesity (for example, *ob/ob* mice) perhaps because this implies a role for active thermogenesis in leanness. The only claim to novelty we can make is the suggestion that brown fat is the major effector of DIT and we see this as a positive development in an established line of inquiry and not a reversal of previously accepted views.

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1. Lotfi, M., Macdonald, I. A. & Stock, M. J. *Br. J. Nutr.* **36,** 305–309 (1976).
2. Macdonald, I. A. thesis, Univ. London (1977).
3. Rothwell, M. J. & Stock, M. J. *Br. J. Nutr.* **41,** 625–628 (1979).
4. Rothwell, N. J. & Stock, M. J. *Nature* **281,** 3–35 (1979).
5. Miller, D. S. in *Animal Models of Obesity* (ed. Festing, M. F. W.) 131–140 (Macmillan, London, 1979).
6. Rothwell, N. J. & Stock, M. J. *Proc. Nutr. Soc.* **39,** 45 (1980).
7. Rothwell, N. J. & Stock, M. J. *Proc. Nutr. Soc.* **39,** 20 (1980).
8. Rothwell, N. J. & Stock, M. J. *J. comp. physiol. Psychol.* **93,** 1024–1034 (1979).
9. Foster, D. O. & Frydman, M. L. *Can. J. Physiol. Pharmac.* **56,** 110–122 (1979).
10. Rothwell, N. J. & Stock, M. J. *J. Physiol.* (in the press).
11. Neumann, R. O. *Archs Hyg.* **45,** 1–87 (1902).

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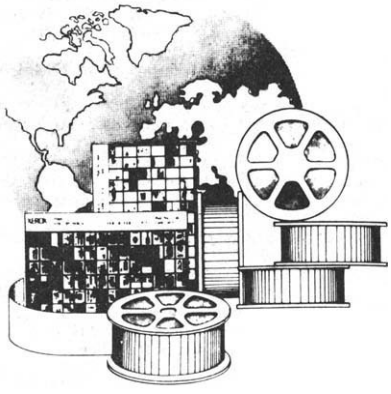

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TEXTBOOK SUPPLEMENT

Self-consciousness and the book for students

Students' books have at least one thing in common with fairy stories: they are written for the use of one section of the population by another. The authors of students' books are people who teach students and who pretend, sometimes accurately, to know what students need to know. Patronizing though this convention may seem, it appears to have survived even the fierce confrontation between students and teachers of the 1960s. The groups of students with ambitions then to help shape the university curriculum, often by the addition of "socially relevant" studies, seemed implicitly to accept that even though what to teach might be up for grabs, how to teach was for teachers to decide. Teachers have been more than willing to follow this prescription. In this convention, the contributors to the symposium of reviews of students' books which follows are teachers not students.

But does not this practice include a dangerous risk of circularity? If teachers write students' books, review them in journals such as *Nature* and also recommend them to their students, is there not a possibility that teachers' preconceptions of what students need will be self-justifying prophecies? At the level of the secondary school, there is ample evidence that this danger is not fanciful. Until the early 1960s, in Europe and North America, most science courses were marked by an almost ostentatious neglect of modern (or postwar) science. Now, some crusty teachers (mostly outside schools) argue, the pendulum has swung too far in the opposite direction. Students at schools learn about the relevance of DNA to human genetics before they know the first thing about the hydrogen bonds that hold these molecules together. Both the old and the new conventions about the content of science courses are indeed determined by teachers, acting as often as members of examination boards or members of curriculum study groups as in the classroom. Fortunately, the dangers are less serious at the level of the university.

The most obvious safeguard is the incoherence of the market in which the would-be authors of students' books must make their way. At some point in a teaching career, the impulse to see a successful course between hard covers, and launched on a wider world than that of the teacher's own university, seems to take hold. Publishers are willing accomplices, in normal times eager to make sure that their own portfolio of standard course books is comprehensive enough for them to offer almost everything to almost anybody (and also to keep their sales forces productively employed). Given only a modicum of independent judgement by teachers who are not themselves the authors of books, there is every reason to hope that the most intellectually successful course books will be those that sell most widely. Even when it seems that the simple influence of the market may be distorted by extraneous considerations — a textbook by a well-known author or from a distinguished institution — there is no serious reason to fear that students' teachers fail to recommend books honestly.

But is it proper that the important task of educating students should be determined in such a haphazard way? Might it not be better that university students, like those in schools, should be provided with books and other teaching materials that are the product of a self-conscious examination of students' needs. In short, should universities borrow from the schools and embark on deliberate curriculum development? In the past decade and more, this challenge has been cheerfully accepted in several places and in many fields of study. Groups of university teachers have spent their weekends hammering out a logical structure for some course of instruction, have gone back to their classrooms to try out the ideas on their students and have in the end subscribed their names to the title pages of one or more books. The result is not always the unsatisfactory compromise that might be expected. Especially when the objective is to devise a teaching course that crosses established disciplinary lines, the product of these endeavours may be stimulating and valuable. If, for example, the objective is to give the teaching of a subject a greater sense of history (a crying need in most fields, especially in mathematics), most practising subject teachers will acknowledge that they could profitably make use of help from others. To the extent that course-books are increasingly the product of several people who work together as a teaching team, some of the more obvious objectives of curriculum development are already assured. Yet formal curriculum development has inevitably hung fire outside the schools. Why should this be?

Some parts of the explanation are self-evident. Good teachers are rightly as suspicious of the all-embracing course-book as their better students are resentful of the intellectual straitjacket it represents. One essential part of a university education is to learn, mostly by hard experience, how to gather different nuggets of understanding from various sources, many of them books. The course-book is thus potentially an enemy of learning, a crutch and not a challenge. Second, those who teach in universities quite properly hold that it is their duty to teach their subjects in distinctive ways. It would be a great misfortune if this principle were eroded, for teaching is at its best a creative process.

These are sufficient reasons why a certain degree of anarchy in the production of students' books is to be welcomed. Another is that many books, originally devised as books for students, have become important components of the scientific literature. Feynman's three-volume *Lectures in Physics* has been for the past twenty years as much a challenge to his peers as to the undergraduates at the California Institute of Technology to whom the lectures were first delivered. In the same field but in a very different style, Max Born's *Atomic Physics* remains a classic of succinctness and a good read, even though the subject has been transformed in the past half century. R. H. Fowler's *Statistical Mechanics,* an enthralling graduate textbook, illustrates a different point; when amended to take more explicit account of the relationship between statistical mechanics and thermodynamics (as *Statistical Thermodynamics,* by Fowler and E. A. Guggenheim), the result was awkward and too didactic. Plainly, self-consciousness can be dangerous.

Self-consciousness is nevertheless not entirely to be outlawed. Indeed, there are many points in the undergraduate curriculum at which there is an urgent need of it. Why is it, for example, that chemists and physicists are still taught thermodynamics as if they were being taught quite different things? Why is the physics of engineering so different from physics as taught in physics departments? Why is the common ground between undergraduate organic chemistry and biochemistry so hard to find? Why are there nearly as many different kinds of "mathematics" as there are universities at which mathematics is taught? Why, throughout the undergraduate teaching of science, do teachers embark apparently independently on the design of experiments and projects, unable to profit from what has been done elsewhere? The dangers of self-consciousness notwithstanding, is it not time that university teachers wrestled more diligently with these problems?

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Mendel meets DNA: the teaching of genetics

D. J. Cove

THE publication in the past year of seven textbooks of genetics, all aimed to provide support for an introductory undergraduate genetics course, provides an opportunity not only for a comparative review of the books but also for a more general examination of how the subject should be taught at this level. The teaching of genetics poses problems both of strategy and of tactics. Strategically, a logical order of development is needed but not easy to devise. The two alternatives most commonly adopted are the classical or "Mendel before molecules" approach and the modern or "Let's start with DNA" approach. But in both of these there is the risk of compartmentalizing genetics and this risk is increased when quantitative, population and ecological genetics are considered as a third discrete topic generally tacked on behind.

The table accompanying this review indicates the gross order adopted in each of the seven texts, but the depressing point to emerge is that although some of the authors say they have opted for an historical approach (e.g. Avers; Burns) and some claim their approach to be modern and molecular (e.g. Levine; Russell), while others put forward their approach as neither historical nor modern but "born of pedagogical logic" (Fristrom and Spieth), there is rather little difference in the end product. The order of presentation is obviously different but there is little feel for the unity of the subject. Because formal, Mendelian genetics was established before biochemical genetics, we are not obliged to teach the two subjects separately; because quantitative and molecular genetics have developed largely independently of one another, the two subjects need not be kept apart for ever; and because the concept of dominance was introduced before the physiology of gene expression was understood, we are not compelled to continue to teach about dominance in an abstract and, to the student, often mysterious manner. There are of course risks in becoming a slave of the integrated approach and the sequential nature of learning inevitably imposes the need to teach some topics before, rather than alongside, others. Nor should it be forgotten that textbooks are not intended, at least in most cases, to be lecture notes for teachers. To be useful as reference works some compartmentation is desirable, but in many cases integration brings not only the advantage of reinforcing the unity of the subject of genetics but also allows some topics to be understood much more easily. The phenomenon of epistasis, for example, presents few problems to the student if introduced in the context of interaction between genes affecting the same biochemical or developmental pathway and this approach minimizes problems encountered by students in distinguishing epistasis from dominance, which are in molecular terms two quite distinct phenomena. It is interesting that Wagner *et al.*, a text which opts for the "Mendel before molecules" approach, treats epistasis in just this way, while Levine, a "Let's start with DNA" book, deals with epistasis without at the same time considering gene function. Curiously, Fristrom and Spieth, despite being a long text, fails to mention epistasis at all.

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Genetics. By C. J. Avers. (Van Nostrand: 1980.) *Modern Genetics.* By F. J. Ayala and J. A. Kiger Jr. (Benjamin/Cummings/Addison-Wesley: 1980.) *The Science of Genetics: An Introduction to Heredity.* 4th Edn. By G. W. Burns. (Macmillan, New York/Collier Macmillan, London: 1980.) *Principles of Genetics.* By J. W. Fristrom and P. T. Spieth. (Chiron/Blackwell Scientific: 1980.) *Biology of the Gene.* 3rd Edn. By L. Levine. (C. V. Mosby: 1980.) *Lecture Notes on Genetics.* By P. J. Russell. (Blackwell Scientific: 1980.) *Introduction to Modern Genetics.* By R. P. Wagner *et al.* (Wiley: 1980.) Further details of all books are given in the table accompanying the review.

IMAGE UNAVAILABLE FOR COPYRIGHT REASONS

Before leaving teaching strategies, I want to return to overall order of presentation. There is no one order in which to teach elementary genetics which is superior or correct. Each teacher or textbook writer will favour his own individual approach. Some will wish to impose an evolutionary bias, others a molecular bias, some will favour a physiological and cellular emphasis, others will regard consideration of the whole organism to be more important. What is vital is to adopt a strategy which emphasizes how each field of genetics is dependent on each other field and how all are interrelated. In this respect I cannot recommend with enthusiasm any of the seven textbooks reviewed here, nor for that matter any other text published previously, although, at least in parts, Avers, Ayala and Kiger, and Wagner *et al.* make the most effort.

And so I come to tactics. It is fortunate that most students find genetics an interesting and stimulating subject since many undoubtedly find it difficult to master. Understanding the logic involved in connecting experimental observation to final deduction is a problem encountered across the whole of science but the connecting thread is especially difficult to follow in many genetical experiments. The logic involved is, however, crucial to genetics and is why the subject remains so powerful in the analysis of living systems. Teaching students to understand this logic so that they can themselves make deductions from data is one of the biggest problems facing the lecturer, who can be forgiven for being driven from time to time to the conclusion that geneticists can only be made in heaven. Here, a textbook can help the student if the relevant logic can be returned to again and again until it falls into place.

Unfortunately, the logical and rigorous way of treating a topic is not necessarily the most interesting and is certainly the most demanding. The device of giving the conclusions first and then showing how the experimental findings are consistent with these is the approach most often adopted, but this is a poor compromise, too easily deceiving the student into believing that he has understood the principles involved. The point can be illustrated by considering the use of deletion mapping to produce fine structure maps of genes. The most straightforward approach to teaching this topic and essentially that used by those of the textbooks under consideration which deal with it, is to take as a starting point an established deletion map, usually out of deference to history the rII genes of phage T4, and to show how point mutants can be located on this map within regions defined by the end points of the deletion mutants. Most students find this approach easy to follow yet, if asked to construct a deletion

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Genetics textbooks — how they compare

	Avers	Ayala and Kiger	Burns	Fristrom and Spieth	Levine	Russell	Wagner *et al.*
Number of pages	659	844	608	657	542	258	573
Text potential (k words)[1]	360	430	320	410	300	190	360
% diagrams/figures/tables[2]	28	50	23	15	24	41	35
Problems	Yes	Yes	Yes	No[3]	Yes	No	Yes
Answers							
Given?	Yes	No[3]	Yes	No[3]	No	No	No
Quality of explanation	Fair	Good[3]	Poor	—	—	—	—
Order of presentation							
DNA before Mendel?	No	No	No	Yes	Yes	Yes	No
Meiosis before Mendel?	No	Yes and no[4]	No	Yes	Yes	Yes	No
Population, quantitative and ecological genetics last?	Yes	Yes	No	Yes	Yes	Yes	Yes
Price — hardback	£17.20	£9.95	£13.95	£11.50	NA[5]	NA	£11.15
	$22.95	$21.95	$19.95	$21.95	NA	NA	$26.20
Price — paperback	NA	NA	£6.95	NA	£12.75	£6.80	£6.45
	NA	NA	NA	NA	$17.95	$19.75	$14.15

[1]Text potential = number of pages of text × estimated maximum number of words which could be included on a full text page. [2]Estimated as the mean of 50 pages chosen at random. [3]Available separately in a "solutions manual". [4]Mitosis and meiosis introduced briefly in an introductory chapter but returned to later. [5]Not available with this binding.

map given raw data, many find the task extremely difficult. Here, a worked example might be helpful. Only Avers, Ayala and Kiger, and Fristrom and Spieth make some attempt at this but their treatments are not really adequate.

I have examined these texts more generally for their treatment of genetic logic, considering not only their approach to Mendelian genetics but also their treatment of experiments such as that of Crick and coworkers which elucidated the general nature of the genetic code and which remains an outstanding example of logical genetical deduction. On these criteria Ayala and Kiger perform best overall and genetic logic is one of Russell's stronger points. Fristrom and Spieth treat Mendelism well (but omit the Crick experiment). The other texts are more patchy; Avers acknowledges the "brilliant reasoning" of Jacob and Monod but does not attempt to duplicate it. Levine, although stressing in the preface the importance of presenting "wherever possible, the experimental procedures and data that have led to our present concepts in genetics", is the most descriptive, making little attempt to present even Mendelism in a logically rigorous way. All the texts, once again, display in their treatment of Mendelism, the extent to which they are constrained by the historical development of the subject. That Mendel was able to make his deductions concerning the mechanism of inheritance, despite the fact that the characters he used were expressed in diploid tissues and that the character differences showed complete dominance, makes him all the more to be admired. It is unnecessary however to expect students to cope with segregation, diploidy and dominance all at one bite. I favour introducing Mendelism by first considering segregation for characters expressed in haploid tissues and dealing next with a "diploid" character in which incomplete or codominance allows the immediate recognition of the 1:2:1 segregation in the F2 generation. This course is not without hazard, with students occasionally attempting to explain a pattern of inheritance in *Drosophila* by proposing that its tissues are haploid. The introduction of diploid segregation by way of incomplete dominance, however, has certainly proved satisfactory.

All the texts adopt the historical approach, in some cases appending incomplete dominance and haploid segregations as embarrassing exceptions, and in passing implying that dominance is an invariant relationship between two alleles rather than dependent on the level and aspect of the phenotype examined. Perhaps more seriously, the texts give the impression, initially, that each gene has only two alleles, one functional, the other mutant, an approach which results in the student having to backtrack when confronted with many topics introduced later. Ayala and Kiger once again perform best when these points are considered. Although describing complete dominance first, incomplete and codominance are introduced in the same paragraph, and the segregation ratios associated with them follow immediately after those obtained with complete dominance; multiple alleles are also introduced early on, but no emphasis is given to the artificiality of the concept of a unique wild-type allele.

Problem solving provides an excellent way in which the student of genetics can satisfy himself that he has understood the ideas which have been introduced to him. Surprisingly, since all seven texts are American, two contain no questions (but see table). Three contain questions but no answers, although in the case of at least Ayala and Kiger these are obtainable as a separate booklet. Burns contains both questions and answers but most of the answers are so brief as to be educationally worthless. Only Avers has, within the single book, questions together with answers which contain at least some explanation, although these are not as good as those provided by Ayala and Kiger in their solutions manual (which, rather curiously, is said in their preface to be "available to the instructor"). I see no point in including questions in a textbook unless answers, containing a clear and adequate explanation, are also readily available. Space is at a premium in all textbooks but in a genetic textbook set problems must have a high priority.

The authors of all seven books have obviously tried to make their books up to date. All contain at least some treatment of recombinant DNA technology and its implications but they do not all deal with intervening non-coding sequences in eukaryote DNA. Both Ayala and Kiger, and Fristrom and Spieth contain good if brief accounts of both these topics.

The texts vary very widely in their emphasis on figures and diagrams. Ayala and Kiger justifiably draw attention to the generous provision of illustrations in their book and must be given the prize not only for the clarity of their figures but also for general layout and design. Fristrom and Spieth is not only the most sparsely illustrated but has a layout which immediately looks dull. This is unfortunate for this textbook is certainly not dull in content. There is little to choose between the other five books with respect to design or their use of figures.

And so I come to where I am no doubt expected to name my best buy. No general textbook covering the whole of genetics can be expected to please everyone; inevitably, each will have strong and weak points. Although not perfect, Ayala and Kiger is, as I have indicated already, obviously superior on a number of counts. Despite its layout, I liked Fristrom and Spieth's treatment of many topics, including developmental genetics, and so I would make this my second choice. Of the rest, I am unsympathetic to the descriptive style of Levine, but others may be less critical; Russell, which is one of a "Lecture Notes on . . . ." series, is too superficial to

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be used as a reference book and does not provide as good value for money as the other books; the remaining three books all have good points — Burns, for example, emphasizes human genetics throughout, Avers integrates the genetics of discontinuous and continuous phenotypic variation, and an important point in favour of Wagner *et al.* is that I have already received favourable reports of it from my own students. However, I am disinclined to recommend any of the books except Ayala and Kiger, and Fristrom and Spieth, except perhaps for reference in libraries. □

Microbial genetics abbreviated

J.R.S. Fincham

Genetics of Microbes. By Brian W. Bainbridge. Pp.193. (Blackie/Halsted: 1980.) Hbk £16.95; pbk £7.95, $27.95.

THIS book deals with fungi as well as with bacteria and bacteriophage, and attempts to take the reader from the fundamentals up to the frontiers of research. It covers a reasonably comprehensive range of topics — some old, such as parasexual reproduction in fungi, and others rather new, including mitochondrial genetics of yeast, genetic analysis in streptomycetes, and *in vitro* manipulation and cloning of DNA in plasmid and phage vectors. The basic systems of gene exchange in bacteria, including transposons, are all described, albeit sketchily in some cases. Lambda phage gets considerable attention.

To cover all this in 170 pages (exclusive of bibliography), as Dr Bainbridge has attempted to do, is an ambitious undertaking, and I cannot say that he has been wholly successful. Conciseness is too often the enemy of clarity. Much information is presented in the form of complex diagrams, often based upon recent reviews and in themselves good and well chosen. But the captions are usually very short and often supported by only a telegraphic sentence or two in the text. Sometimes essential explanations or even facts are omitted in the rush to get on to the next topic. For example, after a brief account of gene conversion we come, a few pages later, to evidence from random meiotic products which is interpreted as favouring the idea of negative crossover interference. No hint is given that, in the light of tetrad analysis, data of this kind can be largely explained in terms of gene conversion. In the same chapter, an otherwise clear account of the Poisson distribution and its application to recombination theory is spoiled by a paragraph implying that a mean chiasma frequency of 0.5 corresponds to 50 per cent recombination (a factor of two missed here). In the chapter on the analysis of the yeast mitochondrial genome, the uninformed reader is not told that mitochondrial recombination is customarily detected and assayed in diploid hybrid clones and he will probably come away with the impression that it happens only in meiosis.

This, then, is not the ideal book, but it has its good points. The account of genetic engineering will be useful for beginners and the discussion of *Streptomyces* genetics provides information not readily available elsewhere except in the specialist literature. The chapter on DNA repair and mutation gives an accurate up-to-date account, albeit virtually confined to effects of UV. Several of the tables and figures provide good summaries of things one often wants to look up, and the bibliography is a useful guide to the primary sources. □

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Genes and behaviour

B. Burnet

Behavioral Genetics: A Primer. By R. Plomin, J. C. DeFries and G. E. McClearn. Pp.417. (W. H. Freeman: 1980.) Hbk £14.80, $24.95; pbk £8.10, $14.95.

BEHAVIOURAL genetics is the discipline linking the biological and behavioural sciences. It is concerned with the systematic study of the heritable bases of behaviour and the role of behaviour in evolution. The problem with teaching it is that intelligent discussion of the interesting issues depends upon a background knowledge of genetics, behaviour, statistics and evolution, and this is what the book provides. Following some historical background about Darwin and Galton, the authors discuss the theory of evolution in relation to behavioural genetics and sociobiology. There is a clear and concise introduction to genes and chromosomes and to the quantitative and population genetic approaches to the study of behaviour. Especially valuable is the careful explanation of how genetic variation in human behaviour is studied and how this yields information about the *environmentality* as well as the *heritability* of behavioural differences. They draw together the results of recent surveys which indicate that IQ differences are less heritable than earlier studies have suggested.

The authors take their material mostly from studies on human beings and small mammals except in their chapter "Mechanisms of Heredity and Behaviour", which includes bacteria, protozoa, round worms and insects. Those who, like myself, believe that invertebrates will for some time provide the arena for testing rival theories about the causal networks linking genes and behaviour may feel that advances using fruitflies and nematodes deserve more emphasis. The assertion on p.41 that "amphibians arose from the invertebrate lobe-finned fish" may discomfort vertebrate palaeontologists, and I note with interest a novel enzyme in Fig.10.1.

This is an excellent text, not only for the quality of the scientific explanations but because it deals critically with several controversial issues. It shows why behavioural genetics is intellectually exciting and practically relevant to the human situation. I recommend it for course work in the biological and behavioural sciences, and to the general scientific reader. □

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"Mini Luria"

D. H. Watson

Introduction to Modern Virology. 2nd Edn. By S.B. Primrose and N.J. Dimmock. Pp.251. (Blackwell Scientific/Halsted: 1980.) Pbk £7.50, $19.95.

IN this second edition of *Introduction to Modern Virology,* forming part of the Blackwell *Basic Microbiology* series, Dr S.B. Primrose has been joined by his colleague Dr N. J. Dimmock. The 251-page volume is amazingly comprehensive, forming a "mini Luria" in that it adopts the same general plan of Luria's *General Virology* (Wiley, 3rd Edn 1978). The treatment follows that given in a number of universities offering comprehensive virology courses as a component of undergraduate microbiology degrees.

An introductory chapter gives an historical survey of the development of ideas on the nature of viruses; a useful guide on basic techniques of animal virology is appended. Chapters on structure and nucleic acids of viruses are followed by six core chapters on the virus growth-cycle. Later chapters deal with such topics as virus pathogenicity, the immune system and interferon, vaccines and chemotherapy, and tumour viruses. A useful addition is a concluding chapter surveying virus families giving their "official" names, although the authors appear to be unaware of the recent U-turn

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of the relevant international committee which is dropping the unwieldy family name "herpetoviridae" in favour of "herpesviridae", after having strenuously resisted the arguments to do so in 1975!

The honours student will inevitably need to refer to more comprehensive volumes and a useful reference list is provided. Nevertheless, most teachers of virology would be more than delighted were their students to have a firm grasp of all the material in this book.

Inevitably, individual teachers will be slightly critical on one or two minor points. I was a little disappointed that the section on neutralization of viruses by antibody, reflecting the inclusion of Della Porta and Westaway in the reference section, appeared to suggest that the multiple critical site theory is fully substantiated. My preference would be to refer students to a more balanced view as represented in the reviews of Mandel. Others, perhaps, may be a little surprised at the omission of any reference to sub-unit vaccines in the relevant chapter. However a book of this size inevitably involves condensation of argument and selection of some topics at the expense of others, and there can be no serious claim of major omissions. The volume is likely to be welcomed by students and teachers alike. □

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Spoiling the story

R. Whittenbury

Microbiology. 3rd Edn. By G. A. Wistreich and M. D. Lechtman. Pp.786 + appendices. (Glencoe, Encino, California/Collier Macmillan, London: 1980.) $20.95, £11.95.

GOOD plays, good novels, even good textbooks for undergraduates all display a common quality — a mastery of the subject, revealed in clarity of style, fluency of writing and in the skilful telling of the tale. *Microbiology,* I'm sorry to say, is not of this class. It is divided into six sections, three on basic principles of microbiology and three on related topics (immunology, medical microbiology and control of micro-organisms, and applied microbiology). Although much of the book is unexceptionable, it is badly flawed particularly in the area of basic principles.

A more personal criticism should be directed towards its "education props" style of presentation. All students (one likes to believe) relish a bit of an intellectual challenge. In this instance the challenge will, I fear, be boredom, rather than comprehension. Each chapter is prefaced by a brief description of the goodies to come and a list of major "take-home" messages, point by point, to be absorbed. In other words, the bones of the plot and the fate of the main characters are revealed before you've even had a chance to clap eyes on the chapter proper, itself sub-sectioned and highlighted. To reinforce this approach the chapters finish with a fairly lengthy sub-sectioned summary, again point by point — "Ribosomes are important in protein synthesis" — a list of review questions and a supplementary reading list (a bit too advanced for the readers the authors have in mind).

Excellent micrographs, black-and-white and colour photographs, and clear tables and diagrams (with one or two notable exceptions) all help to hammer home the message. One quibble I have is the pointless and irritating colour banding of the tables.

The more serious problems, emerging mainly in the basic-principle sections, stem from the remit the authors have set themselves — to write a text for students who have had some exposure to chemistry or biology, but not necessarily to both. These emerge as ill-written paragraphs and sub-sections, many errors of fact, contradictions, and over-simplification of concepts and complex issues. The last sometimes leads to incomprehensible statements or to truisms, some of which even raise spurious queries in the mind of the reader, for example "Phototrophs are noted for their photosynthetic activity". All phototrophs, of course, are photosynthetic organisms, by definition; the very form of this seemingly innocuous statement sows the seed of an idea that there may be some other and more important feature characterizing all phototrophs. Failure to integrate to ensure consistency, if not accuracy, adds to the confusion. For instance, what is written about autotrophs, photosynthesis, methane oxidizers and methane producers contains many examples of all these faults. Many of the errors (typographical included) are really inexcusable in a basic textbook.

Four final examples of the sort of thing you shouldn't find in a work of this nature are: a gross typographical error — "phoyodynnyhriv bacteria"; a fiction — that carbonates can be reduced via the cytochrome-mediated electron transport chain; a contradiction — that lignin is an exception to the rule of microbial infallibility (after giving earlier a list of bacterial and fungal species which break down lignin); a paragraph containing misinformation and needing no valedictory comment from me:

> The microorganisms in the rumen also provide other specific functions, such as the synthesis of amino acids and vitamins. Some microorganisms leave the rumen and are digested in other parts of the gastrointestinal system to serve as a major supply of proteins and vitamins for the ruminant. This is particularly important for the nutrition of ruminants because grasses are deficient in protein.

The authors have much remedial work to do before the fourth edition emerges and before the book, as a whole, can be recommended for use as a student text. Even if the text were faultless — and, potentially, it is a good example of its genre — my preference would still be for the Roger Stanier style (*General Microbiology* by R. Y. Stanier *et al.;* Prentice-Hall/Macmillan, London, 4th Edn 1977), an example of the sort of book I had in mind when writing the opening sentence. □

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Nearly Fungi perfecti

Peter E. Long

Introduction to Modern Mycology. By J.W. Deacon. Pp.197. (Blackwell Scientific/Halsted: 1980.) Flexi £6.50, $18.50. *Introduction to Fungi.* 2nd Edn. By John Webster. Pp.669. (Cambridge University Press: 1980.) Hbk £30, $79.50; pbk £9.95, $21.95.

MYCOLOGY, traditionally a taxonomically orientated part of a botany honours syllabus, now finds a place in microbiology courses where greater emphasis is placed on fungal development and function. *Introduction to Modern Mycology* is a welcome, though belated, addition to Blackwell's *Basic Microbiology* series, reflecting the new approach. Very much a book for beginners, it is a clearly written, compact, explanatory account of fungal structure, growth and development, coupled to consideration of the role of fungi in nature. Taxonomy and morphology are confined to the introduction and there is a concluding chapter on ways of restricting fungal growth. The 14 chapters include an amazing number of topics, aided by lengthy figure captions that impart additional information. The author admits to neglect of some traditional areas; these include the mechanisms of ascospore and basidiospore discharge, while slime moulds receive scant attention. Such omissions are largely excusable, but I draw the line at his failure to make any mention of the lichens, which number roughly a quarter of all described fungal species. The illustrations are not so well chosen as the contents and sometimes appear crude, especially in

The Great Gray Owl: Phantom of the Northern Forest, reviewed in *Nature* **288**, 519, 1980, will be published in the UK, Europe and the Middle East in March of this year by Oxford University Press. Price will be £9.95. The book is available in the USA from The Smithsonian Institution or through bookshops.

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comparison with the next volume. It is a pity that the selection of further reading, mostly other books, is so restricted since it will not satisfy the interest in fungi that the book will arouse. Dr Deacon's book is currently the best, short, undergraduate introduction to fungi on the market, albeit at the cost of some oversimplification.

The second, expanded, edition of *Introduction to Fungi* is, by contrast, a reference work, suitable for second- or third-year undergraduates and of great value to all mycologists. It retains the format, style and superb standard of illustration found in its predecessor. Thus it creates the impression of a traditional mycology textbook. This is deceptive, as within its taxonomic framework lies an enormous amount of information on fungal structure, cytology, genetics, physiology and behaviour, as well as other general topics. All the subject matter is presented with precision and detail backed by innumerable references to original sources. The pre-existing chapters have been thoroughly revised, the treatment of slime moulds has been greatly extended and there are two new and welcome chapters on the general attributes of the Eumycota and on the Fungi Imperfecti. More attention than before has been paid to the production of the book, resulting in better placed and printed illustrations, a clearer hierarchy of headings, and more obvious stressing of terms through use of bold type instead of italics. In all, it is a considerable improvement on the first edition.

However, instructors who have to decide between recommending this book and the latest edition of Alexpoulos' *Introductory Mycology* (Wiley, 2nd Edn 1979) are placed in an unenviable position. Professor Webster undoubtedly provides a greater body of information about the fungi, in a work that is as helpful in the laboratory as it is in a study. Unfortunately the book is neither so easy to read nor apparently so topical as its American counterpart. This is largely a matter of presentation. *Introduction to Fungi* has only seven long chapters, necessitating numerous subheadings. In some cases it is not clear where a particular section ends. For example, "Breeding for resistance" (p.509) seems to continue into a discussion on rust life-cycles. Students will also encounter difficulties, at times, in sifting essentials from the mass of ancillary information as key points are not always well emphasized.

Any book of this length and quality, for which the author also takes on the onerous task of illustrator, carries the risk of falling just short of perfection. The present volume comes very near to escaping this fate. Despite its drawbacks for novices, it will undoubtedly and deservedly enjoy a wide market and will be regarded by many as a classic.

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Plant taxonomy surveyed

J.G. Vaughan

Plant Taxonomy and Biosystematics. By Clive A. Stace. Pp.279. (Edward Arnold/University Park Press: 1980.) Flexi £8.95, $29.50.

PLANT taxonomy in the modern context is very much a multidisciplinary subject and encompasses various types of botanical investigations and techniques. According to the author, this book is a broad survey of the subject and is aimed primarily at university undergraduate students and others in similar establishments. The information presented is largely limited to green plants, with the emphasis on angiosperms; fungi and lichens are excluded.

The first section deals with the scope of taxonomy and the types and development of systems of classification, the second with the sources (structure, chemistry, chromosomes, breeding systems, plant geography, ecology) of taxonomic information. The book is rounded off with discussion of the process and mechanics of classification. The treatment of the subject is thus fairly comprehensive and the text is well supported by references. Bearing in mind that the book is intended for undergraduates, I do have some minor criticisms— Fig. 3.7, showing 31 types of stomatal complex, could be daunting to such students, and the method of double-diffusion serology illustrated in Fig. 4.3 is not typical of that used by chemotaxonomists. Also, the distinction between lemon and orange on skin colour (p.225) would not be significant in West Africa, and possibly some other areas.

Stace's book is an important contribution to the literature of plant taxonomy in that it deals with all the classical and modern facets of the subject in a concise and readable fashion. Undergraduates, after a basic training in plant science, will find it most useful. Indeed, it is a valuable work for any scientist or institution seriously interested in plant variation.

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Transport for botanists

James F. Sutcliffe

Transport in Plants. By U. Lüttge and N. Higinbotham. Pp.468. (Springer-Verlag: 1979.) DM58, $30.50.

RECENT research on the transport of materials in plants has been reviewed extensively in the first three volumes of a new series of the *Encyclopedia of Plant Physiology*, published by Springer-Verlag in 1975–1976. The same publishers have now issued a further book on the subject, based on Professor Lüttge's *Stofftransport der Pflanzen* (1973). It is a concise, authoritative and up-to-date account of research in a field to which the authors themselves have made important contributions.

Inevitably, in a book of moderate size covering a wide subject, some topics receive less detailed treatment than others. In general, the material reflects the research interests of the authors and so there is extensive coverage of membrane transport and its relationship to metabolism. On the other hand, phloem transport is disposed of in about ten pages and transport of water receives hardly any attention.

The first part of the book is devoted mainly to a straightforward account of various physical phenomena associated with transport processes, including osmosis, electrochemical potentials and electrogenic pumps. The Nernst equation, Goldman voltage equation and Onsager coefficients are explained. The presentation is clear and the mathematics involved should not be beyond the comprehension of a physiologically-minded biologist.

Part II is concerned with transport into cells and begins with a summary of current views on the structure of cell walls as a prelude to consideration of the concept of free space. This is followed by an account of the structure of membranes and various ideas on membrane-transport. The authors then turn to the complexities of the multicompartmented, vacuolated plant cell. Some background knowledge of cytoplasm is assumed. There is a useful appraisal of the value and limitations of the kinetic approach to the analysis of ion fluxes and an impartial assessment of the concept of dual mechanisms.

In Part III the control of transport and its relationship to metabolic processes are considered. Particular attention is paid to the role of photosynthesis in regulating ion-fluxes in algal cells and in isolated chloroplasts, and there is an interesting chapter on the influence of phytochrome and growth-regulating substances on membrane transport.

The final section of the book deals with inter-cellular and long-distance transport within plants, and covers such topics as the movement of ions across the root cortex, functioning of glands, ion-relations of stomata and transport in sieve tubes. Non-botanists will find this part of the book

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difficult to follow if they have no previous knowledge of plant structure and some simple line-diagrams illustrating the salient features would have been helpful.

The book is attractively produced and, in all, a useful volume. I shall recommend it enthusiastically to advanced undergraduates embarking on courses in plant physiology and to their teachers.

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World of botany

Peter D. Moore

Vegetation of the Earth (and Ecological Systems of the Geo-biosphere). 2nd Edn. By Heinrich Walter. Pp.274. (Springer-Verlag: 1979.) DM 29.40, $15.40. *Plant Geography.* 2nd Edn. By Martin C. Kellman. Pp.181. (Methuen: 1980.) Hbk £10; pbk £4.95, $10.95.

I AM sure that all ecologists and biogeographers will welcome a second edition of Heinrich Walter's *Vegetation of the Earth*. Perhaps the two most valuable features of the first English edition were its simplicity and clarity of terminology and arrangement, and its liberal use of physiological data, so well selected and integrated with climatic and distributional information. Although this new edition is slightly expanded, these two features do not appear to have been substantially strengthened. Clarity has, in my opinion, not been enhanced by the introduction of a new system of jargon. We now have eubiomes, zonobiomes, orobiomes and pedobiomes, where once we had deserts, savannahs and forests. The salt desert of Utah is now a eubiome of the pedohalobiome of zonobiome VII. Such jargon rivals terms like "dedesertification"! On the physiological side, the book remains rich in useful examples, but some omissions are conspicuous. I find it surprising that space is not found for a discussion of C_4 photosynthesis and its intriguing biogeographical implications; crassulacean acid metabolism is given only a few lines.

On the positive side, the book's expansion now permits coverage of geographic areas neglected in the first edition. This will be particularly welcomed by Australians and New Zealanders, who now receive more than passing mention — an unusual feature in any biogeography book. Overall, Heinrich Walter's book remains the best, condensed account of world vegetation on the market.

Martin Kellman's second edition of *Plant Geography* has been extensively rewritten and the material substantially reorganized. Three themes permeate the book: plant population biology, environmental history and evolution. It is a difficult task to bring together such extensive subjects at an undergraduate level and to attempt their integration. This is especially so if one feels it is necessary to start from a very basic level of biological background; within half a page he has defined and discussed mitosis, meiosis, polyploidy and introgressive hybridization. Fortunately, more resources are allocated to survivorship curves, seed demography and r and K selection. Species interactions and responses to the environment are considered, but here the accounts suffer from a lack of physiological information. The development of the interaction theme through coexistence to plant community analysis is logical, but there follows an unconformity, with chapters on techniques and then the human factor being slipped in as brief appendices.

Kellman aims in this book to stimulate in the minds of his readers questions relating to the evolution of plants and plant distributions. His careful selection of interesting topics should enable him to achieve this end, and his arrangement of material has certainly resulted in the second edition being a considerable improvement on the first.

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How to stimulate students of plant physiology

L. J. Audus

Plant Hormones and Plant Development. By W. P. Jacobs. Pp.339. (Cambridge University Press: 1979.) Hbk £21.50, $16.95; pbk £7.25. *Biochemistry and Physiology of Plant Hormones.* By T. C. Moore. Pp.274. (Springer-Verlag: 1979.) DM 49, $27.

THE advent of extremely sensitive and chemospecific assay techniques has revolutionized plant hormone studies in the past two decades, and has brought into question many time-honoured theories of the hormone control of plant growth and development. What, then, do I look for in books such as these, intended as they are for undergraduate students of plant biology? Apart from a critical and balanced assessment of the present state of our understanding, I would hope to find that kind of stimulating presentation and analysis that would fire the student to go further into this fundamental science.

Of the two books under review, only Jacobs's comes anywhere near to meeting these requirements. He believes, and rightly so, that it is essential to emphasize "the development of research ideas" and this he has done quite admirably by taking carefully selected phenomena with which he himself has been most actively concerned (vascular development, hormone transport, abscission, flowering, for example), and making exhaustive, rigorous, critical yet constructive assessments based on his well-known PESIGS rules for evaluating the hormone evidence. Such treatment makes exciting reading which is marred only by occasional lapses into somewhat facetious and unnecessarily caustic criticism of other workers' deficiencies. It is, on his own admission, a very personal treatment, as witness for example the inclusion of data from his own researches in more than half of the plentiful illustrations in the text. Biochemical matters are virtually absent and the highly selective treatment leaves some important phenomena untouched (for example, geotropism) and the cover of the various hormone actions much restricted (for gibberellins only the action in flowering is adequately presented). Regrettably, ethylene is regarded as not being a hormone.

On the other hand Moore's book is predominantly biochemical, even molecular-biological. While Jacobs presents his chapters under phenomena, Moore does so under individual hormones, thus largely omitting the important matter of hormone interactions. Developmental responses are briefly covered but few (for example, dormancy) are treated satisfactorily. Detailed consideration of phytochrome, although systematic and complete, seems to be out of place in this book, especially since the role of hormones in flowering is so over-condensed and incomplete as to be dangerously misleading to the student. On the whole, hormone biochemistry is well treated; for example there is, among others, a nicely balanced appraisal of modern ideas of auxin mechanism.

While Jacobs is rigorously honest in the matter of priorities, Moore is occasionally slack; in fact his whole approach to references is puzzling. References to key researches are often not given in the lists at the end of each chapter, although these lists are much swollen by references to works which are not mentioned anywhere in the text. The student will not find this helpful.

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Of the two, then, Jacobs's book would be my choice for recommendation to students, although it still falls short, in my opinion, of the earlier book by K. V. Thimann *(Hormone Action in the Whole Life of Plants;* University of Massachusetts Press, 1977) which fulfils all Jacobs's criteria concerning the development of ideas. It is just as rigorous, objective and stimulating but is better in that it maintains just about the right balance between molecular biology and developmental physiology. □

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Biological "wheres"

Paul A. Colinvaux

Biogeography: An Ecological and Evolutionary Approach. 3rd Edn. By C. B. Cox and P. D. Moore. Pp.234. (Blackwell Scientific/Halsted: 1980.) £7.50, $19.95. *Biogeography.* By E. C. Pielou. Pp.351. (Wiley-Interscience: 1980.) £13.30, $31.30.

THESE are two books about the new biogeography, one a survey for undergraduates with little background, and the other a provocative series of essays touching on the state of the art.

Biogeography was the founder subject of both evolution and ecology. In Alfred Russel Wallace's *The Geographic Distribution of Animals* of 1876, evolution was triumphant and Sclater's six zoogeographic realms, each full of animals partly evolved in isolation, became evidence for evolutionary theory. The circumpolar zones of Allen, which recognized ecological realities — boreal forest, temperate forests, rain forests in rings of latitude on every continent regardless of ancestry — were ridiculed by Wallace as the study of mere adaptation. The Sclater–Wallace classification lasted until fairly recently, through to the plant geography of Good. In the intervening years, ecologists almost avoided the word "biogeography" and talked instead of life in the great formations, roughly corresponding to Allen's old zones, later to be known by Clements' unnecessary and regrettable term, biome.

But with ecology taken away, biogeography could be little more than the study of relationships towards the goal of a better phylogeny. True, a speculative fringe found excitement in continental drift and land bridges, but they made few geological converts.

For half a century, evolutionary biogeographers on an Earth of fixed continents proceeded to fill in details. At the last, Darlington in *Zoogeography* (Wiley, 1957) told us of endless radiations from the heartland of Africa (the Old World Tropics), perhaps feeling the ghostly presence of a rejected Gondwanaland.

Then came renewal. Remanent magnetism and plate tectonics gave us back Wegener's drifting continents; Pangaea and Gondwanaland lived. But even more vibrant was the incursion of ecology, as theorists of the Hutchinsonian school taught us to look at colonization, dispersal and speciation as ecological processes on a geographical scale. MacArthur and Wilson gave us *The Theory of Island Biogeography* (Princeton University Press, 1967).

Cox and Moore, in their third edition, press on with their successful look at this new biogeography through ecologists' eyes. Not only does the species equilibrium on islands stand with drifting continents as a central theme, but prime ecological concepts like Lindeman efficiency, niche and resource-partitioning find their places also. A possible worry is that ecological concepts that really require a mastery of competition theory have to be handled with the logistic hypothesis and its derivatives left out — competition is taken for granted rather than debated. Yet Cox and Moore make a brave show of giving modern ecology to students with no training in it, all the same.

Pielou's book is a survey by a master ecologist to be used by, as she puts it, "Senior undergraduates, and interested biologists and earth scientists of all kinds". She says that she is "probably rash" to attempt such a thing, but she was not. She has done what she said she would so well that no scientist of a related discipline can now be excused ignorance of the possibilities of the new biogeography.

Each of the ten chapters of Pielou's work will stand by itself as an essay, so you can really begin to read the book where you like. I recommend starting with the third, "Evolution, Phylogeny, and Geographic Spread". This sounds conventional enough; the title could be in a book from the 1930s. But you will instantly be drawn into a splendid analysis of modern systematics controversy that I just could not stop reading, even though in bed with 'flu at the time. Have you wondered what was behind the guerrilla war between Halstead and the Natural History Museum (*Nature* **288**, 208; 1980)? — read Pielou on cladistics, phylogeny and the evidence of biogeography.

She is a little hard on geologists who would not accept the biogeographic evidence for continental drift before the advent of plate tectonics — the honest truth was that it did not seem parsimonious to move a continent in order to accommodate a fossil. She puts island biogeography somewhere in the middle of the book, as a subject appealing to sectional interests, then follows that by a chapter on "Geographical Ecology". From then on the book wields the Hutchinsonian ideas of the evolutionary play in the ecological theatre, until we bow out with thoughts on the fundamental mechanisms of polyploidy and gene flow in the last chapter. It is a book both to read and refer to, highly modern but based on profound understanding of the history of an ancient subject. Many a graduate student will have both thoughts and work shaped by it. □

Paul A. Colinvaux is a Professor in the Department of Zoology at The Ohio State University.

Understanding ecosystems

I. J. Linn

Ecology: A Textbook. By H. Remmert. Pp.289. (Springer-Verlag: 1980.) Pbk DM 37, $21.90. *Ecology.* 2nd Edn. By R. E. Ricklefs. Pp.966. (Chiron/Nelson: 1979.) Hbk $22.95, £20; pbk £12.50. *Principles of Ecology.* By R. Brewer. (Holt, Rinehart & Winston/Holt Saunders: 1979.) $22.95, £10.50.

WHEN I was a student, many years ago, my early ecological diet was firmly founded in L. A. Borradaile's *Animal and its Environment* (Frowde, 1923) and A. S. Pearse's *Animal Ecology* (McGraw-Hill, 1926). Both of these books devoted much space to a detailed examination of the fascinating ways in which animals are adapted to survive in a wide range of habitats — the sort of thing which nowadays *Scientific American* does rather well. It is pleasing to note that in the new edition of Robert Ricklefs' standard work, despite some changes, he continues to open his argument with a solidly evolutionary approach to the adaptation of organisms to their physical and biotic environment. Of course he goes far beyond that — much has happened in half a century — and for me Ricklefs' *Ecology* is still the basic ecological reference book that no biologist (ecologist or not) can afford to be without. The jargon is all there; if you are not sure what is meant by "character displacement", or of the precise difference between "exploitation" and "interference" competition, the lucid explanations in text and glossary will help you out. For the numerate, there is liberal use of the necessary descriptive equations; and for those, like myself, whose command of matters mathematical is somewhat patchy, there is help in succinct English.

There are faults in the book, of course,

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but by and large they are minor. The major weakness is that, while the author by no means ignores work and examples from outside the Americas, his concentration on matters American (no doubt deliberate) amounts to a regrettable bias, not only for his readership outside America, but also for his domestic readers, who would benefit from broader horizons. As I scan my own bookshelves, I see that the great majority of books on ecology in English produced in the last 20 years or so are American, so it is a particular pleasure to welcome Hermann Remmert's *Ecology: A Textbook*, in which many European and some African examples are found alongside the American ones.

Remmert's book is only a third of the size of Ricklefs', so the treatment of the subject is bound to differ. Remmert has decided to concentrate on autecology, population ecology and ecosystems, and this approach is largely successful. The style of the book is also very different. You will find no jargon here, and very few equations, only plain and most readable English (the translation from the German by Dr Marguerite Biederman-Thorson is excellent). The result is a first-rate exposition of many of the basic concepts of ecology which can be read with profit by biologist and non-biologist alike. Professor Remmert is very concerned that the magnitude of the task of understanding complex ecosystems should not be underestimated, and he is sceptical of the value of mathematical models. He also has some harsh words for what he calls "ecological quacks" who offer advice on how to improve mankind's lot, and he points out that the need is to maintain in good condition the environment to which we are so well adapted, and that improvement in this context is a meaningless concept.

For this reason, presumably, he has included no section on applied ecology, unlike Richard Brewer, whose *Principles of Ecology* is otherwise similar in size and coverage, though with the expected American bias. Brewer, from his comments on Spaceship Earth, is equally concerned to conserve essentials, and his sections on the ways in which the ecologist can go about offering sensible advice seem to me not unreasonable, though I doubt if they would satisfy Professor Remmert. □

I. J. Linn is a Senior Lecturer in the Department of Biological Sciences of the University of Exeter.

Aquatic biology, past and future

C. M. Yonge

Fundamentals of Aquatic Ecosystems. Edited by R. S. K. Barnes and K. H. Mann. Pp.229. (Blackwell Scientific: 1980.) Flexi £7.80, $22.50. *The Ecology of Streams and Rivers. Studies in Biology,* 122. By Colin R. Townsend. Pp.68. (Edward Arnold: 1980.) Flexi £2.10.

WHEN I was on the staff of the Plymouth Laboratory the basic methods for the quantitative determination of phosphates and nitrates in the sea were being elaborated, while, at the same time, a descriptive shore ecology was being supported by experiments revealing such unsurprising facts as that organisms highest on the shore are most resistant to the effects of desiccation. Certain of us were making tentative advances in comparative physiology.

These two admirably presented and illustrated books are almost entirely concerned with the results of half-a-century of subsequent progress. The properties of the ecosystem emerge as no less significant than those of the organism or even populations of organisms. Energy flow is revealed as all-important.

The more comprehensive *Fundamentals of Aquatic Ecosystems* is the ably edited work of ten authors, each highly qualified to deal with some aspect of the subject which includes the increasing human exploitation, by aquaculture, of the products of marine and fresh waters. No better introductory survey of these highly significant aspects of modern biology could be desired, one certain to fire the imagination of senior pupils and first-year undergraduates. How rewarding, surely, to teach students already so well informed.

Relevance to the future arises out of comment in the book on the recent discovery near the Galapagos Islands of unique animal communities around submarine volcanic vents at depths below the penetration of light. Consequent description of the sulphur cycle, on which this life depends, involves mention of the industrial production of sulphur dioxide and its influence on the basic chemistry of the Earth. Such effects on the biosphere emerge as among the major hazards of coming decades.

The smaller *Ecology of Streams and Rivers* serves almost as a supplement, concerned as it is with the maintenance of populations within running waters with the constant danger of being carried seaward to destruction. It is an excellent addition to the *Studies in Biology* produced by the Institute of Biology. □

Sir Maurice Yonge is an Honorary Fellow in Zoology at the University of Edinburgh.

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Concepts of animal behaviour

Aubrey Manning

Introduction to Ethology. By K. Immelmann. Pp.237. (Plenum: 1980.) $22.50, £14.18.

IMMELMANN defines ethology as "the study of the biology of behaviour". This covers almost everything, but whilst I am no longer convinced that ethology as a separate field exists, it remains easy to recognize the influence of the ethological *approach* in all branches of behavioural science. Certainly, we need textbooks which introduce ethological concepts, particularly as so many students are now plunged directly into elaborate sociobiological arguments and tend to give short shrift to the study of an individual's behavioural organization and development, which has always been ethology's strong point. The translation of Immelmann's book is going to be useful for it covers a lot of ground.

My heart sank at first when I saw, yet again, illustrations of Tinbergen's herring-gull bill patterns and stickleback models. Yet there are also plenty of fresh examples and it is healthy to have many from the German literature. Anyway there is nothing to say that old examples (or old concepts) are not still the best, provided that, where necessary, they are re-interpreted in the light of modern thinking. I do feel Immelmann falls short here in some cases, for example when describing the effects of the hawk-goose silhouette on game birds or when discussing terms like "action-specific energy" as causal factors in behaviour.

From a student's point of view the book is clearly set out, with sensible use of sub-headings. Systematization is taken too far in places, with Immelmann's excessive use of formal terms to classify his material. Some of these, such as "ethoendocrinology" and "acquired releasing mechanism (ARM)", are unlikely to be encountered again outside the pages of this book.

I would have liked more examples to be spelt out in detail, but Immelmann has a large field to cover and in general he summarizes very clearly. The chapter on social behaviour is particularly valuable as a modern re-statement of the ethological approach with due emphasis on sexual behaviour and care of the young. In summary, a good book to work through with students on an introductory animal behaviour course. □

Aubrey Manning is Professor of Natural History at the University of Edinburgh, where he works on and teaches animal behaviour. He is author of An Introduction to Animal Behaviour (*Edward Arnold, 3rd Edn, 1979*).

More neuroscience

Trevor W. Robbins

Behavioral Neuroscience: An Introduction. By C. W. Cotman and J. L. McGaugh. Pp.838. (Academic: 1980.) $21.95, £13.80. *Physiological Psychology.* By T. S. Brown and P. M. Wallace. Pp.634. (Academic: 1980.) $20.95, £11.20. *Fundamentals of Human Neuropsychology.* By B. Kolb and I. Q. Whishaw. Pp.501. (W. H. Freeman: 1980.) $17.95, £8.95.

"HOW could anyone know enough to write a whole textbook?" Brown and Wallace thus modestly quote one of their own students in the preface to *Physiological Psychology.* Indeed, the sometimes dazzling technical advances in neuroscience have led to a proliferation in knowledge which defies the capacity of individual writers to achieve the degree of synthesis or balance required of a text. Brown and Wallace's book, however, is but one in a long line of such attempts and so it seems reasonable to consider some criteria by which textbooks in this area can be assessed.

One of my own criteria is to ask whether it is possible to guess from the relative qualities of the various sections, the specific research interests of the authors. I concluded (partly correctly) from *Physiological Psychology* that one or other of its authors had special expertise in the chemical senses, sexual motivation or circadian rhythms, since these sections seemed the most detailed and best-explained.

Most of the other topics covered fit into the familiar organization of chapters on introductory neuroanatomy and neurophysiology, sensory systems, motor systems, neural plasticity, learning and memory, and "higher processes". These subjects are generally covered adequately, but suffer from temptations seldom avoided by textbook writers in physiological psychology.

The first of these is to act essentially as an uncritical, passive reporter of what are considered to be main issues or experiments. For instance, Brown and Wallace's discussion of hunger and satiety flits uneasily between the vagaries of "central" and "peripheral" theories, for example, "But as we shall see later in this section, the pendulum is swinging back again, and is now somewhere in the middle" (p.291). Now this may be an accurate assessment of the state of that particular art but this lack of intellectual guidance would not instil me, nor I suspect, an undergraduate, with much confidence.

A second, related temptation is to avoid such shows of indecision by skirting any real discussion of the questions posed, or of the nature of the underlying processes. Thus, an otherwise tidy account of "brain stimulation reward" is marred by a meagre half-page of theoretical discussion of

Primates in the lecture theatre

W.C. McGrew

Social Behaviour in Primates. By Neil Chalmers. Pp.256. (Edward Arnold/University Park Press: 1980.) Flexi £6.50, $19.95.

THE rise of behavioural studies of non-human primates over the past two decades has stimulated a concomitant growth of university courses, especially in North America. Because of their cross-disciplinary nature, such courses are taught in departments of anthropology, biology, and psychology, thus creating a large market for textbooks. The first wave of such books appeared in the early 1970s, Jolly's *Evolution of Primate Behavior* (Macmillan, 1972) and Rowell's *Social Behaviour of Monkeys* (Penguin, 1972), for example, but these are obsolescent.

Now comes a new entry into the area, written by an English primatologist experienced in African field-studies. The substance of the book is in six chapters — on groups, social structure, infant development, sex, dominance and adaptiveness. It focuses on fieldwork but calls in laboratory studies when appropriate, for example on hormonal effects. The approach is selective rather than encyclopaedic, and one of the author's strengths is his skill in choosing apt examples. The most serious drawback is that consideration of the function of behaviour is postponed until the final quarter of the text. What precedes is solid discussion of the causation of behaviour, but until the end there are no hooks upon which the reader can hang the obvious questions which this discussion stimulates. For example, why should primates live in groups, unlike many other mammals? Why do some, but not all, primates breed seasonally?

Still, these are but quibbles, as Chalmers has produced an admirably lucid textbook which deserves to be widely used. □

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Twelve years after the appearance of the second edition, Cambridge University Press have published a new edition of *Excursion Flora of the British Isles* by A.R. Clapham, T.G. Tutin and E.F. Warburg. The new edition is available with a plastic cover and costs £12.50.

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incentive motivation, coming almost as an afterthought. Sadly, this approach encourages the present tendency of "behavioural neuroscience" to be a subject driven by phenomena rather than by thought.

A third temptation is to litter the text with trendy material, perhaps not only to capture the excitement of contemporary research but also to make the opposition obsolete. Brown and Wallace have rightly introduced much interesting recent material, for example, on enkephalins and pain, the neurochemical substrates of learning, and, impressively, on "brain grafts". However, the "shock of the new" is not always right. What is exciting today may be reinterpreted or not even replicated, tomorrow. Although it is perhaps healthy for students to hear of controversy, it is dangerous to present it as fact. In addition, driven by the thirst for novelty, the reader expects updating in every topic, whereas Brown and Wallace's account of dopamine levels in schizophrenic brain neglects important work.

These are perhaps harsh criticisms of what is a well-produced and solid textbook. The choice and clarity of the figures is particularly good, and the writing emulates in readability that of Carlson's *Physiology of Behavior* (Allyn & Bacon: 1977), although in my opinion the latter is a more successful textbook.

In contrast to *Physiological Psychology*, Cotman and McGaugh's work represents a significant change of style in textbooks on the subject. Although much of the material overlaps with the former book, the title *Behavioral Neuroscience* emphasizes its preoccupation with neurobiology, which I found to be the strongest feature of the book.

The authors, while compiling a vast fund of useful information, have attempted to solve the problem of its critical assessment by enlisting about 20 specialist collaborators. Unfortunately, despite this expertise, the book fails to achieve that essential thematic coherence and balance in level of presentation. Some of the chapters, such as those on learning and memory are very detailed, whereas others, for example on thirst and hunger, are not. The superior colliculus is barely mentioned in the chapters "Seeing" and "Moving", and neuropsychological topics such as functions of association cortex and frontal lobe, and aphasia, are given short shrift.

The book is excellently produced with one glaring exception; the many sketches accompanying the text are poor substitutes for scientific drawings, and in a few cases, are embarrassing and superfluous.

Can there then be an ideal textbook in physiological psychology? In preparing my own courses I have found none, although many of the textbooks are very useful. If we eschew the possible unfortunate development of textbooks being written (and reviewed by) committees, then a better approach would seem to build a modular course by using books on specialist topics, such as R. Melzack's excellent and inexpensive *The Puzzle of Pain* (Penguin, 1973).

Superficial inspection of Kolb and Whishaw's *Human Neuropsychology* suggests that it will be a valuable weapon for the latter armoury, especially since it covers areas only poorly represented in the other books reviewed. I was impressed with the attempt to integrate both theory and human and animal experiments in sufficient depth to provide adequate coverage of subjects such as the neural mechanisms of language.

Although this book will be invaluable for advanced courses in physiological psychology and clinical psychology, I doubt it will be the last word on textbooks in neuropsychology. However, I also doubt that we have seen the last textbook of physiological psychology. □

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More sight than sound

Stuart Sutherland

Perception: The World Transformed. By L. Kaufman. Pp.416. (Oxford University Press: 1979.) Hbk £10.75; pbk £6.95, $14.95. *Perception: An Applied Approach.* By W. Schiff. Pp.493. (Houghton Mifflin: 1980) $18.50, £9.95. *The Psychology of Visual Perception.* 2nd Edn. By R.N. Haber and M. Hershenson. Pp.431. (Holt, Rinehart & Winston/Holt Saunders: 1980.) $22.95, £10.50.

TWENTY years ago there was no worthwhile textbook on visual perception. This deficit was removed by the publication in 1975 of Kaufman's *Sight and Mind* and Rock's *An Introduction to Perception*, the former an advanced-level undergraduate textbook, the latter rather more elementary, but both excellent. We now have three further textbooks on vision — despite their titles, *Perception: The World Transformed* and *Perception: An Applied Approach* deal almost entirely with vision and both treat audition in a cursory and inadequate fashion.

Of the three new books, Kaufman's is the easiest to read, but is less up to date and is considerably more elementary than the other two. Haber and Hershenson's *The Psychology of Visual Perception* is exceptionally lucid and gives fair and unbiased accounts of the theories with which it deals. Schiff's text is both the least conventional and the most scholarly: as well as summarizing our current knowledge of perception, it shows how this knowledge can be applied to such problems as the design of aids for the blind and the legibility of text, and its extensive references provide the reader with ready access to the recent literature on vision.

The books have different virtues, but they all share the same fault. The mechanisms of visual perception are the way they are because they have evolved to carry out a particular task, namely, to reconstruct from the fragmentary two-dimensional data in the retinal image a three-dimensional model of the environment and to recognize the objects represented in that model. Although all the authors acknowledge that vision has a function, they fail to relate the details of the research they describe to that function. Thus, they describe the Gestalt psychologists' laws of Prägnanz (for example, the tendency to group together similar visual elements arranged in a straight line or smooth curve) without giving any indication of how the application of these laws enables the visual system to carry out its primary function.

Moreover, none of the books gives any account of the computer models that have been devised over the past decade to extract three-dimensional representations from a visual image. The attempt to construct such models has revealed the nature of the

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problems that the visual system must solve, and in many instances has suggested how it actually does solve them. Not one of the books makes any reference to the work of the late David Marr, perhaps the only genius to work on the psychology of vision since Helmholtz himself. Marr's work in fact started where Helmholtz stopped: Helmholtz rightly insisted that visual perception was mediated by a series of complex inferential processes, but he was unable to specify with any rigour what these processes were. The existence of the digital computer has allowed Marr and his associates to construct computational models of the visual system that have thrown a completely new light on how it must function. Unfortunately, his work is not easy to follow and too few psychologists have grappled with it: the only good popular account is to be found in J.P. Frisby's *Seeing: Illusion, Brain and Mind.*

Two of the publishers of the three books under review have served their authors badly. In *Perception: The World Transformed* some of the figures are unnecessarily hard to interpret (e.g. p.128), others have incorrect legends (e.g. p.190), and at least one name that appears in the index does not appear on the specified page or indeed anywhere else in the text. The sins of Oxford University Press are, however, comparatively venial compared with those of Houghton Mifflin. *Perception: An Applied Approach* is the worst designed book I have ever encountered. It can be read only with extreme difficulty since each page contains three columns of text with no justification of the right-hand margins; much of the information purveyed is contained in those horrid boxes so popular with American publishers, but Houghton Mifflin have plumbed new depths of idiocy by spreading a single box over as many as five different pages, with areas of normal text and figures spattered randomly around its edges; as though these faults did not make the reader's task sufficiently difficult, the book's designer has made it almost impossible to associate the figure legends with the relevant figure, even going to the length of placing figure and legend on different and non-facing pages. The format of the book is a triumph of bad design, a nice irony given that the book's content is intended to show how a knowledge of vision can improve the design of artefacts.

In summary, then, *Perception: The World Transformed* is an excellent introduction for the novice; *The Psychology of Visual Perception* is a good but conventional treatment and is nicely complemented by *Perception: An Applied Approach*, which includes much material not normally found in undergraduate textbooks. □

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Information in mind

Angus Gellatly

Cognitive Psychology. By R.L. Solso. Pp.499. (Harcourt Brace Jovanovich: 1980.) £10.95, $17.95. *Cognitive Psychology and Its Implications.* By J.R. Anderson. Pp.503. (W. H. Freeman: 1980.) £8.10, $15.

IN THE preface to his book, Robert Solso identifies two key questions raised by the information-processing approach to cognition: what are the stages through which information is processed? and in what form is information represented in the human mind? Although each of these texts addresses both questions, they differ in the emphasis given to them.

Anderson's major concern is the representation of knowledge, and much of his book is an exposition of recent models of long-term memory that employ a propositional format; indeed, he sees such models as the major achievement of modern cognitive psychology. The organization of the book reflects his commitment; peripheral stages of perception and attention are covered in a single 40-page chapter, after which interest turns to the evidence on knowledge representation. Only when his conclusions on this issue have been established does Anderson move on to apply them to the perennial problems of learning, memory, reasoning and language. By contrast, Solso's commitment is more to the methods of information-processing psychology than to a particular theoretical stance within that framework. His is the more traditional approach, starting with the detection of sensory signals and charting the flow of information into the processing system. Over 100 pages are devoted to reception and early analysis of stimuli, with consideration of memory in its varied manifestations forming the second part of the book, and higher-order cognitions the third. Both authors display an admirable familiarity with the vast literature. Anderson's clearly defined viewpoint lends his book a vigorous proselytizing air and a unity of purpose, but Solso, by allowing greater space to studies published earlier than the past decade, suggests a longer-term perspective. Anderson provides particularly good chapters on mental imagery, schemas, cognitive skills and deductive reasoning. Solso is also good on mental imagery and on attention, and he scores over Anderson by including a chapter on cognitive development.

On the debit side, the two books are open to a number of the same criticisms. Both are concerned almost exclusively with what cognitive psychologists do in the laboratory using undergraduate subjects and computers, rather than with cognition in any broader sense. Neither author makes more than passing reference to relevant neuropsychological findings, to the issues of individual and cultural differences, or to applied studies of reading. Perhaps, granted the assumptions they make, these are allowable omissions, but both authors advance staggering claims for future developments and applications of cognitive psychology that are in stark contrast to the well-worn examples of such applications that they can cite.

For teaching purposes, what are the relative merits of the two books? Solso perhaps provides the more general and easier introduction to cognitive psychology, Anderson is for the second- or third-level option course. □

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Entomological ethology

Stewart Evans

Introduction to Insect Behavior. By M. D. Atkins. Pp.237. (Macmillan, New York/Collier Macmillan, London: 1980.) Pbk $9.95, £6.50.

ALTHOUGH the value of studies in invertebrate behaviour is widely accepted, these animals still tend to be neglected in textbooks of ethology. Some general texts, such as those by Alcock (*Animal Behavior: An Evolutionary Approach;* Sinauer, 1975) and Manning (*An Introduction to Animal Behaviour;* Edward Arnold, 3rd Edn, 1979), do strike a satisfactory balance between different animal groups but others give the impression that almost all of the important research is on birds and mammals.

Clearly, students need access to a wider coverage of the behaviour of invertebrates, and Atkins's book goes a long way towards providing this for insects. In an admirably clear account, he concentrates on functional aspects of behaviour and, with well-chosen examples, illustrates both its complexity and variability. I believe that undergraduates will enjoy reading the book; they will find it informative, up-to-date and, in parts, dynamic. For instance, the chapter which includes a consideration of genetic aspects of behaviour underlines the way in which both traditional ethological techniques, such as those employed in Rothenbuhler's studies of hygienic and unhygienic honey bees, and entirely new ones, such as those involving the use of *Drosophila* mosaics, are making

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inroads in this area of research.

One major disappointment is, however, that the author almost totally ignores the work of insect neurophysiologists. It is difficult to understand how Huber's research, or that of several others in the same field, could be overlooked. Students ought to be aware of this work and, in my view, the book suffers from missing the opportunity to include it. This omission apart, I think that the book is good value for money. It is attractively illustrated and provides a good reading list; hopefully students will be stimulated to make extensive use of it. □

Stewart Evans is a Senior Lecturer in the Department of Zoology at the University of Newcastle upon Tyne.

Diversity of insects

R. W. Thorp

Entomology. By Cedric Gillott. Pp. 729. (Plenum: 1980.) Hbk £31.19, $49.50; pbk £14.18, $22.50.

THIS book is a comprehensive introductory college text for a senior undergraduate class in general entomology. It is designed to contrast with the "traditional taxonomic approach" which provides little information on insect physiology and ecology.

The 24 chapters are grouped into four major divisions: evolution and diversity; anatomy and physiology; reproduction and development; and ecology. The first of these comprises about 45% of the book, and deals with insect diversity, arthropod evolution, and classification and identification of insects, and includes a chapter on external structure. A key for the identification of all orders is provided; description of each order is followed by discussions of structure, life history and habits, and phylogeny and classification, including brief mention of the more common or important taxa. Most of the rest of the book, divisions 2 and 3, deals with structure and function of the integument, and all internal systems, and with embryonic and post-embryonic development. Finally, the author considers how insects relate to their abiotic and biotic environments, and concludes with a chapter on applied entomology entitled "Insects and Man".

The strength of the book is in the areas of physiology , developmental biology and the ecological relationships of these to abiotic environmental factors. Also, the chapters on arthropod evolution, insect diversity, classification, and identification at and above the ordinal level, especially the phylogenetic discussions, are more complete than in most single-authored texts.

Weaknesses include the lack of a discrete section on insect behaviour, and inadequate discussions of population dynamics and life tables, forest entomology, zoogeography and community ecology, thermoregulation in insects, interactive effects of abiotic factors, host specificity, economic thresholds, and methods for insect population management. The section on chemical control is poor in that possible resistance to insect growth regulators is not discussed, nor are the potential uses of anti-juvenile hormone compounds, antifeeding compounds and chemosterilants. The absence of keys and descriptions of infra-ordinal taxa reflects the author's feeling that identification is best relegated to laboratory exercises and to collections made by the students; however, this means that an additional text with keys to and descriptions of families will be needed for the lab.

It may be concluded from the author's criticism of introductory texts which are taxonomically oriented, and from my criticisms of the gaps in his coverage, that it is not easy to find a suitable text to accompany both lectures and practical work in entomology. Although most authors agree that a balanced coverage is needed, one is quickly overwhelmed by the vast amount of material to be covered in a quarter or semester. After the fundamentals of the subject have been covered, there is often little time left to introduce the fascinating diversity of natural history data available. Thus, the "balance" achieved in the introductory texts and courses currently available seems to differ according to the bias of the author or instructor. No two texts ever strike the same balance, but perhaps this is not as important as the development of student interest in this fascinating field. This new text by Dr Gillott succeeds in accomplishing this and is one of the better books available for an introductory entomology course. □

R. W. Thorp is Professor of Entomology at the University of California, Davis.

A new "Life" for vertebrates

Barry Cox

Vertebrate Life. By W.N. McFarland, F.H. Pough, T.J. Cade and J.B. Heiser. Pp.875. (Macmillan, New York/Collier Macmillan, London: 1979.) Hbk $29.95, £12.95; pbk £6.95.

THIS book fills a very great gap in the books that one can recommend to students. There are several adequate, sound books on the comparative morphology of vertebrates, and reference to them is certainly necessary if the student is to understand the details of the evidence of their structural and adaptive radiation. Nevertheless, with their lengthy exposition of how the hip bone is connected to the thigh bone, or of the position of the metaconule, such books provide little stimulation to the enquiring mind. Instead, they encourage the poorer student's tendency to cram facts as an alternative to the more demanding activity of trying to understand principles and to solve problems.

As the authors explain in their preface, this book is built around two themes; the evolution and the ecology of vertebrates, dealt with in a way that also integrates relevant aspects of their physiology, behaviour, locomotion or morphology. So although the list of chapter titles includes the familiar groups in the normal phylogenetic order, many other topics are either covered in separate sections within the chapters or make up independent chapters.

The book is divided into 22 chapters, half of which are by a single author (Cade or Pough), while 9 were written by McFarland and Heiser. The chapters seem uniformly to be really up to date in their approach and in the references given at the end of each. An average of only eight references per chapter is rather low, though some others are given after the illustration captions. The illustrations themselves are excellent and well chosen. Many of them have been redrawn, to provide a pleasing similarity of style thoughout the book. In some cases the authors have enlarged on a particular topic (for instance the evolution of amphibian vertebral structure) by means of a lengthy commentary-caption, so that the flow of the main text is uninterrupted.

After a brief introduction to the basic body plan of the vertebrates, the second chapter discusses their ancestry and environment of origin. The third includes very useful sections on the theory of classification and its application to vertebrates, and on species and speciation. The chapters on the different vertebrate groups deal with extinct as well as with the living groups, and with such additional topics as the origin of land vertebrates, adaptations to life in the deep sea, the ecology and physiology of dinosaurs, and the waves of extinction at the end of the Cretaceous and during the Pleistocene Ice Ages. The authors are careful to emphasize that each group possesses abilities and adaptations in its own right and that, for example, ectothermy has positive value in some ways of life rather than merely being a characteristic of groups that "failed" to become endothermal.

Five chapters scattered through the book give useful summaries of the positions of the continents and of the climatic regimes at different periods of the Earth's history. Other chapters deal with the influence that life in the water has had on basic vertebrate

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functions, and on the adaptations of endotherms to rigorous habitats.

In addition to being well-written and well illustrated, the paperback edition is extremely good value. It should put new life into any vertebrate course, and it thoroughly deserves to capture the bulk of the market for vertebrate textbooks. □

Barry Cox is Professor of Zoology at King's College, University of London.

Fossils the French way

John C. W. Cope

Elements of Palaeontology. By Claude Babin. Translated by N. Orriss and edited by D. Palmer. Pp.446. (Wiley: 1980.) Hbk £19, $57; pbk £7.95, $22.50.

IN HIS introduction to this book the editor mentions the important point, certainly not implicit in the title, that there is an "assumption that the reader is not a novice to palaeontological and biological terminology and technicalities". The approach therefore differs considerably from Rhona Black's *The Elements of Palaeontology* (Cambridge University Press, 1970), a book which has rapidly become the standard introductory palaeontological text.

To the British student the first part of the book will prove to be the most useful. The first chapters cover fossilization, fossil collection and preparation and the basics of palaeontological taxonomy. These are adequate treatments of the subject, but the exhortation to "gather the greatest possible number of samples" will not be welcomed by those with an eye on conservation. There follows a useful résumé of palaeoecological methods; this is well illustrated and many examples will be familiar to English-speaking students.

Part Two of the book carries the curious title "Some Facts of Evolution". After consideration of the origin of life, various aspects of fossil plants and some invertebrate fossil groups are considered. Although the treatment is claimed to be selective and not morphological or taxonomic, I find it difficult to understand why major groups such as sponges, bryozoa or gastropods are omitted whilst minor groups such as hyolithids, conulariids and tentaculitids are discussed. Treatment of major groups is often idiosyncratic. Under "Echinoderms", for instance, a short discussion of pentaradiate symmetry is followed by a presentation of the stylophoran/calcichordate controversy. There is no mention of the other 15 or so echinoderm classes — several of which could provide first-rate examples of evolutionary trends. Similarly, treatment of bivalves is restricted to their origins and their Ordovician diversification. The same approach is applied to other groups, an approach which, rather than stimulating, frustrates by its inadequacies. The final chapter in this section contains some 60 pages on aspects of vertebrate evolution, including a substantial section on hominid evolution.

The final part of the book, "Palaeontology and Evolution", briefly discusses some ideas on evolutionary theory.

The translation (from French) is acceptable but rarely more; it contains a few glaring errors (for instance that the bivalve ligament closes — instead of opens — the shell) but peculiar phraseology and word-order frequently make it awkward to read. Even with an English dictionary to hand, a few sentences are totally incomprehensible.

The good illustrations and useful reference lists cannot compensate for the sketchy and disjointed coverage in Part Two of the book, and although it contains much that is interesting, I must say that I found this book a disappointment. □

John C. W. Cope is Senior Lecturer in Geology at University College, Swansea.

Micropalaeontological survey

Alan Lord

Microfossils. By M. D. Brasier. Pp.193. (George, Allen & Unwin: 1980.) Hbk £12, $27.50; pbk £6.50, $14.95.

THE growth and development of micropalaeontology has been closely connected with interest in the world's oceans, coupled with its application to hydrocarbon exploration. The results of the *Challenger* Expedition (1873–1876) stimulated the study of small organisms and their role in modern sedimentation. Stratigraphical studies of fossil counterparts by such pioneers as T. R. Jones in Britain eventually led to the recognition in the 1920s of the use of microfossils for correlation, particularly between boreholes where drilling usually destroyed macrofossils. Since that time various groups of microfossil have been found to be useful for biostratigraphical and palaeoenvironmental purposes, usually with industry recognizing the value of particular fossils in particular situations after initial academic study. The continuing exploration of the ocean floors by D/V *Glomar Challenger* has depended on microfossils for stratigraphical control.

Dr Brasier's book provides a broad survey of microfossil groups, giving information for each on biology, hard- and soft-part morphology, ecology and distribution, relationship to sedimentation, classification and applications, together with suggestions as to further reading and hints for study. Coverage of each group tends to reflect its past importance to geologists — foraminifera, ostracods, spores and pollen, conodonts, dinoflagellates and coccoliths are given space in that order. Also included are a brief introduction, a useful appendix on techniques and a good basic bibliography. The style is economic, without a great burden of cited literature, but with many pointers to the student for further information and enough named examples to round out each chapter. Numerous line drawings illustrate the book; use of these is clearly a matter of personal preference when excellent photographs of microfossils are available nowadays.

The book is thoroughly recommended for undergraduates and junior postgraduates, not least because it complements the more detailed *Introduction to Marine Micropaleontology,* edited by B. U. Haq and A. Boersma (Elsevier, 1978). The paperback is certainly a good buy for anyone with an interest in or need to know about microfossils. □

Alan Lord is a Lecturer in Geology at University College London and teaches part of the MSc course given in the Postgraduate Unit of Micropalaeontology.

Riches of biology

E. C. Cox and R. M. May

Biological Science. 3rd Edn. By W. T. Keeton. Pp.1080 + glossary and index, pp.A51. (W. W. Norton: 1980.) $19.95, £11.50. Study guide $4.95, £2.50; teacher's manual no charge.

TEACHING introductory biology is not easy. An average class has an extraordinarily diverse set of interests, preparations, aptitudes and appetites for the subject. It is never clear, moreover, where one should start and where, or at what level, one should end, given both the richness of the subject matter and the diversity of the audience. These difficulties surface in the pages of many introductory texts, often reflecting all manner of confusions on the part of the author about pace, depth, coverage and readership. The problems are variously solved by dealing only with animals or plants; by reducing everything to a few weak bonds, permuted; by centring the discussion on the fate of mankind; or by omitting, except *en*

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passant, most of the evolution of structure and function by natural selection. Each of these narrow strategies, when vigorously pursued, can appeal to a particular audience. There is no doubt, for example, that a student already well prepared in the natural sciences can, upon arrival at a university, understand and benefit from Watson's *Molecular Biology of the Gene* or Lehninger's *Biochemistry,* two texts normally used by upperclassmen. But there are few such students. Most of us need, and some even want, a broad and responsible introduction to biology taught in a serious way at a high level. Fortunately for us, Keeton's third edition is now available.

There are many features of the new edition that set it apart from its competitors and represent improvements over the second edition. Keeton's approach has always been to begin with the rudiments of chemistry, proceed to the structure and function of macromolecules, cell structure and function, and thence on through behaviour, genetics and evolution, community ecology, the origins of life and the description of the phyla. He covers each topic thoroughly, judging the depth and breadth with care, and adding the proper notes of scholarly caution and scepticism. The book has an air of authority; too many introductory texts (and lecturers) substitute glib metaphor for real explanation (see our earlier reviews in *Nature* **284**, 106; 1980). The text consequently does not always make for a good read, but since it is aimed mainly at students in the natural sciences, and at those graduate students (and their teachers) reviewing areas outside their special competence, this cannot be faulted. No one could be expected to remember everything in this book; indeed, Keeton says in the introduction that he obviously could not remember it all himself.

The broad theme running through Keeton is that evolutionary theory accounts for adaptive strategies. It is the thread that ties together the material on the cell, energy transformation, the biology of whole organisms and, of course, genetics, evolution, behaviour and population biology. This not only gives students some feeling for the deep structure of the discipline but, as important for a beginner, supplies the outline of an image whose shape is finally supplied by what at first sight is an incoherent body of detail, often alarming in its variety.

The prose is enhanced enormously by first-rate illustrations and photographs, full colour as well as black-and-white. Many examples could be given. Two that stick in our minds are the drawings combined with electron micrographs to illustrate typical plant and animal cells with pleasing clarity, and the beautiful colour photographs of the world's major biomes and of plant pollinators in the act.

Inserts on special topics are also used; this is now a fairly standard pedagogical ploy, but it is nonetheless effective, for it allows small essays on advanced or special topics that would otherwise interrupt the text.

A list of test questions and a teacher's manual accompany the text. The former, in our view, is useful but not exceptional. The latter we can recommend, especially to lecturers who have not previously taken on an introductory course. It has several strengths, not the least being excellent advice on how to pose examination problems and on which of the areas covered in the lectures most often confuse the students.

Keeton's untimely death last year was a sad loss. This excellent text is likely to stand as a memorial for some time. □

Edward C. Cox is Chairman of, and Robert M. May Professor in, the Biology Department at Princeton University, where they jointly teach an introductory biology course.

Maths applied

P. T. Saunders

Statistics and Experimental Design. 2nd Edn. By G. M. Clarke. Pp.188. (Edward Arnold/University Park Press: 1980.) Flexi £6.50, $24.50. *Mathematics and Statistics for the Bio-Sciences.* By G. Easen, C. W. Coles, and G. Gettinby. Pp.578. (Ellis Horwood: 1980.) Hbk £25, $90; pbk £8.50. *Statistics for Biologists.* By D. J. Finney. Pp.165. (Chapman and Hall/Methuen: 1980.) £3.75, $9.95.

WHEN the first edition of *Statistics and Experimental Design* appeared, *Nature's* reviewer wrote that it was "clear and well written and may be strongly recommended to non-mathematical students requiring a common-sense grounding in statistical methods". I entirely agree with this judgement. The explanations are lucid, and well illustrated with examples. There are many exercises, for which model solutions are provided. The author has also added a liberal sprinkling of comments which contribute towards an understanding of the methods he is describing and which should help the reader to avoid some of the pitfalls that await the unwary user of statistics.

The contents are not much changed from the first edition. They include an introduction to probability theory, tests of significance (both parametric and non-parametric), correlation and regression, the principles of experimental design, and an account of factorial experiments including corrections for missing observations and non-normality. Students and research workers who can master the material in this not-too-large book should be well equipped to begin using statistics; those who cannot might be better advised to leave statistics alone.

Statistics for Biologists covers a similar range of material, but deals with fewer topics. This is partly because it is shorter, and partly because the author is more concerned with introducing the principles of statistics than with providing instruction in how it is used.

To some extent he succeeds, but I feel he often devotes to comment and discussion space which could more profitably have been used for techniques, even with his stated aim in mind. For example, regression occupies over four pages, yet the author neither explains the principle of least squares nor gives the formula for fitting even a straight line. Surely this must reduce the chance that the student will understand regression, if only because he cannot be given numerical examples to work out.

As an indication of the level to which students are taken, the author writes that the distinction between one- and two-tail tests is "logically rather subtle" and suggests that the reader should ask a statistician to explain it to him later on. I would have thought that this was one of the basic principles that students ought to learn; it is even hard to read many statistical tables if you don't understand this point. The problem does not arise within the book, as no full tables are provided. Students who find the author's style to their taste will profit from reading this book, but it will leave them with a lot more to learn.

If *Mathematics and Statistics for the Bio-Sciences* had been better written, it could have helped to satisfy a real need. But there are too many things wrong with it. The authors have chosen to write in a rather formal style, which they do not always handle well. I was not impressed with their explanations of such important ideas as limits and derivatives; these are difficult concepts for beginners to grasp, and require a better and more carefully worded treatment than they receive here.

The presentation is often slipshod. For example, while it is certainly true that "Two matrices **A**, **B** can only be equal to each other if they are the same size, say $m \times n$, and all their corresponding elements are equal", the authors should have made it clear that this is the *definition* of equality, and that there is not, as their statement implies, some further condition to be satisfied. It also seems odd to introduce the Poisson distribution and even calculate its mean and variance without mentioning the assumptions on which it is based and (therefore) its main application.

The one really good feature of the book is the large number of realistic biological examples and exercises. Anyone teaching mathematics to biologists will find these very useful. But I cannot recommend the book to students, either for classroom work or for individual study. □

P. T. Saunders is a Lecturer in Mathematics at Queen Elizabeth College, University of London.

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Across the span of biophysical chemistry

Joseph E. Coleman

Biophysical Chemistry, in 3 parts. By C. R. Cantor and P. R. Schimmel. Pp.1,370. (W. H. Freeman: 1980.) Pt I hbk £17.70, $31.95; pbk £9.20, $16.95. Pt II hbk £20.70, $37.95; pbk £10.60, $19.95. Pt III hbk £22.40, $39.95; pbk £11.90, $21.95.

IT IS difficult to give a simple definition of biophysical chemistry. Scientists who have considered themselves biophysicists have covered such a wide variety of subjects, from the physics of ion transport to crystallography, that it is difficult to settle on a satisfactory definition. To avoid narrowness, it is perhaps best to define the field as that in which physical methods are applied to the study of biological systems — molecules as well as larger aggregates. To date, such an immense number of physical techniques have been used in this way that it has been difficult to find a textbook which covers even a representative number of them. At the same time, the study of these methods must be coupled to a reasonably complete understanding of the structure and function of the objects under study — in most instances biological macromolecules.

Cantor and Schimmel have chosen to attack the problem of the breadth of biophysical chemistry by producing a three-volume work of nearly 1,400 pages. The concept is admirable. The first volume, entitled *The Conformation of Biological Macromolecules,* sets the stage by discussing in detail the structures to be studied by various physical methods. Here, the authors have in fact set one of the few limits to their scope by confining the discussion for the most part to homogeneous macromolecules. While the existence of macroassemblies is mentioned in appropriate places, this limitation seems reasonable in that most of the best illustrations of the applications of biophysical chemistry come from studies of homogeneous biological macromolecules. The authors on occasion indicate the power of the methods of studying subcellular particles or even whole cells, but this aspect is not emphasized. Hence the first volume is devoted primarily to discussion of protein and nucleic acid structure, polysaccharides, lipids and membranes being given rather less space. The treatment is thorough, starting with composition and chemistry of the constituent monomers and extending to the static structure of the polymers and on to a discussion of the forces that determine protein folding and nucleic acid structure.

Having set the stage by presenting the structures to be studied, the second volume, *Techniques for the Study of Biological Structure and Function,* presents the physical techniques required to determine the macromolecular structures. Since the methods covered are those used to derive the information in the first volume, a purist might quibble about the relationship of the cart to the horse, but as a learning format such an approach has an advantage, since the context for the application of these (bio)physical methods is clear beforehand. The number of physical techniques applied to one or another biological macromolecule has become so great that it is difficult to be all-inclusive, and Vol. II is not. In a number of specific areas the presentation is extensive, as for all types of optical spectroscopy; hydrodynamic techniques are also well covered. Approximately 130 pages are devoted to X-ray crystallography, including scattering, and to a short mention of extended X-ray absorption edge spectroscopy. This is a thorough and well-presented discussion.

In the magnetic resonance section, both NMR and EPR are covered. The EPR discussion is rather brief; no examples of transition metal EPR spectra are given and the student will have difficulty in understanding even the most basic aspects of the interpretation of EPR spectra from the material given. On the other hand, the NMR section gives an excellent presentation of the theoretical and basic aspects, including relaxation mechanisms. The applications section is not as good. Most of the spectra are out of date; NMR technology has improved so much that even ^{1}H spectra from the early 1970s do not do justice to the field.

The third volume is entitled *The Behavior of Biological Macromolecules.* The idea behind this section would appear to be that since the student now knows his structure and the methods for discovering it, he can now use this information to understand some of the more complex interactions of biological molecules. These include ligand binding to macromolecules (both equilibrium and kinetic aspects), the variety of "allosteric" phenomena involved in biological regulation, and conformational equilibria found in proteins, nucleic acids and membranes. Many specific examples of the various methods dealt with in Vol. II are given in Vol. III and make the discussion of applications of biophysical techniques more complete. This last volume also covers some other methods such as differential scanning calorimetry — an important addition. The extensive ^{1}H-NMR studies of t-RNA are presented here with spectra somewhat more up to date than those appearing in the NMR section proper.

The book is sufficiently mathematical to be relatively rigorous without overdoing it. There is an appendix giving a brief review of matrix algebra, and appropriate problems are found at the end of many of the chapters, with answers in the back of each book.

Minor criticisms aside, one has to conclude that the three-volume concept of presentation was well-founded and that the execution is excellent. I believe that this will be the textbook of choice for a course in biophysical chemistry; I can think of no other text that is as complete or as consistently well done over such a broad spectrum of subjects. □

Joseph E. Coleman is Professor of Molecular Biophysics and Biochemistry at Yale University.

Biochemical quartet

David Saggerson

Biochemistry: A Case Oriented Approach. 3rd Edn. By R. Montgomery *et al.* Pp.711. (C.V. Mosby: 1980.) Pbk £11.50, $19.95. *Biological Chemistry: The Molecular Approach to Biological Systems.* By K.E. Suckling and C.J. Suckling. Pp.381. (Cambridge University Press: 1980.) Hbk £25, $59.50; pbk £9.95, $19.95. *Biochemistry and Physiology of the Cell: An Introductory Text.* 2nd Edn. By N.A. Edwards and K.A. Hassall. Pp.448. (McGraw-Hill: 1980.) Hbk £15.95, $38.20; pbk £7.95, $19. *A Guidebook to Biochemistry.* 4th Edn. By M. Yudkin and R. Offord. Pp.261. (Cambridge University Press: 1980.) Hbk £18, $53.50; pbk £6.95, $15.95.

OVER the past 10 to 15 years, many excellent biochemistry textbooks have appeared. To succeed in the face of such competition, any new book must be outstanding or have some unique and unusual features.

The general textbook by Montgomery *et al.* fits both of these criteria. As its title and the sketches of clinicians at work on its cover imply, it is aimed to serve the needs of students of medicine or other health-related subjects. Each chapter first presents a particular set of topics in much the same way as other, more conventional textbooks of biochemistry. This is then followed by discussion of various clinical conditions, questions and problems. The number and range of conditions covered in the book is impressive. The order of chapters is rather unconventional, starting with nutrition, followed by proteins and enzymes and the acid–base balance, before discussion of cellular metabolism and its regulation. It is explained that the introduction of these particular topics early in the book permits a wider range and fuller discussion of clinical cases in subsequent chapters. I cannot argue with this. The book is imaginative, full of information,

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well referenced (up to 1979 in some cases) and clearly set out. As someone who is personally involved with the challenge of imparting biochemical knowledge to medical students in an interesting and relevant fashion, books of this kind are most welcome. It is likely, though, that the book requires a course to be deliberately constructed around it (as the authors do at the University of Iowa College of Medicine).

The Sucklings' book falls clearly in to two parts. The first two-thirds is largely devoted to explaining in chemical mechanistic terms a wide range of biochemical reactions. Since there are few books with this approach, this is useful either to explain biochemistry to chemists or chemistry to biochemists. Each chapter contains a number of problems which are solved with brief explanation at the end of the book; however, it was not always apparent to me that the information presented was sufficient to enable a student to tackle the problems satisfactorily. The last third of the book covers a number of topics such as enzymes as catalysts, enzyme mechanisms (using kinase-catalysed reactions as examples), control of enzyme activity, biochemical techniques (separation methods and structure determination), membrane structure, drugs (design and metabolism) and communication between cells (hormones and the immune response). It is not clear why these topics have been selected for inclusion considering the nature of the earlier part of the book. This curious mixture is presented in a rather superficial way. One cannot but feel that the book would have been better if this last third had been omitted and the topics left for the general textbooks where they are better covered.

Cellular Biochemistry and Physiology by Edwards and Hassall (McGraw-Hill, 1971) has now metamorphosed into *Biochemistry and Physiology of the Cell*. This is similar in size to its predecessor, having the same objectives presented in the same style. In a modest-sized book the authors have managed to incorporate a considerable amount of material and present it in a clear and concise manner. I see this book as being a valuable introductory text for biochemistry students or as a main text for those following biochemistry as a subsidiary subject. The main areas of the subject are dealt with — protein chemistry, enzymes and their kinetics, intermediary metabolism and protein synthesis, together with specialist sections on nerve, vision and muscle contraction — and throughout the book a number of important techniques are introduced and described. I found the discussion of the control of metabolism, including hormones, which has been considerably expanded since the 1971 edition, to be particularly good. As the authors themselves say, increases in knowledge rendering a book "more advanced" may in fact make it easier to read. Incidentally, this book was the only one of the four reviewed here presenting a credible, but simple explanation of the chemiosmotic theory of oxidative phosphorylation.

The book by Yudkin and Offord is the most elementary of the four. In general it presents a superficial outline of biochemistry under the four section headings, "Structure and Function of Macromolecules", "Metabolism", "Molecular Genetics and Protein Synthesis", and "Compartmentation and Regulation". With 250 rather small pages it cannot cover the same amount of ground as Edwards and Hassall; in fact some important topics are missed out and the explanation of others is rather thin. The book also suffers from a rather dull format. I was particularly disappointed to find no discussion of any of the important techniques of biochemistry nor any simple enzyme kinetics; this latter omission makes explanation of some aspects of enzyme action difficult. I also feel that the authors' approach to intermediary metabolism would have benefited from the use of more diagrams and from an attempt to present it within a physiological framework. Metabolism is much more interesting if metabolic specialization of different tissues is explained alongside the physiological conditions which favour particular metabolic processes. □

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Molecular cytology

John Edwards

Cell and Molecular Biology. 7th Edn. By E. D. P. De Robertis and E. M. F. De Robertis, Jr. (Holt, Rinehart & Winston/Holt Saunders: 1980.) $24.95, £10.95.

Is cell biology a discipline in its own right? What, for example, are the prospects for a biological degree course which cuts horizontally across the traditional divisions between animals, plants and microbes, as does biochemistry, but focussed at the level of the cell? Textbooks with "cell biology" in their titles often are heavily weighted towards biochemistry, usually shorn of its tougher shadings into physical and organic chemistry. Not so with this volume, evolved over more than 30 years from a text of general cytology.

The core of this substantially rewritten edition continues to treat the cytoplasmic organelles and nuclei of eukaryotic cells, bringing together ultrastructural, cell fractionation and functional investigations, and is well illustrated and usefully documented (more than a third of the references post-date the previous edition). Mainstream molecular biology (one-sixth of the whole) leads via cell differentiation

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MOLECULAR ELECTRONIC STRUCTURES
An Introduction*

Carl J. Ballhausen
University of Copenhagen

Harry B. Gray
California Institute of Technology

An introductory account of the electronic structures of atoms and molecules with emphasis on the nature of the chemical bonding in various types of molecules. After completing this text the physical chemistry student will be able to handle polyatomic inorganic and organic molecules as well as the usual diatomics and triatomics. The book is useful also for advanced inorganic chemistry courses and as a self-study unit for the chemist who needs to keep current with this rapidly moving area of modern chemistry.

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November 1980, 144 pp., illus., 5½" × 8¼"
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to chapters on muscle and nerve cells.

Integration of cellular and molecular matters could be improved in places; for example, discussion of self-assembly is divorced from some of its best examples, the G/F transitions of filaments and tubules, and there seems to be no hint of the possible role of glycosylation in intracellular traffic. There is a moderate if uneven emphasis on experimental evidence. For example, we meet puromycin in relation to protein synthesis, but not EGTA in excitation-contraction coupling. Though X-ray diffraction appears amongst instrumental methods, the task of explaining the importance of low-angle diffraction by intact muscle is avoided. Further there seem to be rather too many small oddities, even for a volume with so wide a sweep: EDTA works in one experiment by "binding to phosphate groups"; freeze fracture may "disclose the outer or inner surface of a membrane or may even split the membrane lengthwise". Worst, in context of cancer "normal cultured cells generally divide every 24 hours as long as they float freely in the medium". This last points to a wider weakness: of the two main tools of cell biology today, the electron microscope and cell-culture, the authors are clearly more at home with the former. Otherwise they would not propagate the myth of contact inhibition of mitosis, subsume the locomotion of tissue cells with that of amoebae, or omit all but the vaguest reference to fibronectin. The indexing could usefully be three times denser.

Nevertheless, though this book looks more successfully down the hierarchy than up, it does show that teaching centred on the cell (rather than the animal or enzyme) need not lack coherence or excitement. For such a course it would be useful but far from definitive. □

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Textbook evolution

K. Murray

DNA Replication. By Arthur Kornberg. Pp.724. (W.H. Freeman: 1980.) £19.20, $34.95.

IF ONE regards *DNA Replication* as a second edition of *DNA Synthesis,* one should read the two side by side. One then sees a remarkably concise summary of the advances in nucleic acid enzymology that have occurred in the five years that separate the two volumes, and a lucid integration of the role and interrelations of several newly discovered enzymes in the complex and intricate processes whereby genetic information is maintained and utilized. The evolution of *DNA Replication* from *DNA Synthesis* has involved a number of deletions, an appreciable amount of rearrangement and, not surprisingly since it is nearly twice the size of its progenitor, numerous insertions that confer a clear advantage upon the descendant.

The introductory chapters on the structure and functions of DNA and the biosynthesis of DNA precursors are expanded appreciably, while the chapters on DNA polymerase I of *E. coli* and other prokaryotic DNA polymerases remain largely as the classic accounts that they were, which I believe should still be read by all students of nucleic acid biochemistry — incidentally I am glad that one of my favourite quotations of Kornberg remains: "Failure to detect an enzyme activity in a cell-free extract means only that." (p.160).

It is in the discussion of eukaryotic DNA polymerases in Chapter 6 that the expansion and reorganization of material becomes really apparent. The many enzymes involved in various phases of DNA replication are now allotted individual chapters and this, in my opinion, improves the subsequent discussion of replication mechanisms and their regulation. The chapter on bacterial DNA viruses is appropriately updated and expanded, and animal viruses are similarly treated in a new chapter of their own. As before, chapters on DNA repair, recombination and restriction (and now transformation also) and on the synthesis of genes and chromosomes conclude the book. The final chapter is completely revised and introduces modern advances in DNA sequence determination and recombination DNA technology, topics that merit a book of their own. It is, as one expected, an excellent book written with clarity and an economy for transferring information which makes small bacterial viruses look extravagant, and with the authority of one who has done much to lay the foundations of the subject and set standards for others to emulate.

Like *DNA Synthesis, DNA Replication* will be widely valued both as text and as a reference book. It should be on the shopping list of all students in biological subjects — and chemists too, for that matter, in this era of biotechnology — but it will be equally valuable to research workers. If it stimulates the need for a third version, I am sure Kornberg will feel suitably rewarded. □

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Biomembranes — agreement and discord

Dennis Chapman

Biological Membranes: Their Structure and Function. 2nd Edn. By R. Harrison and G. G. Lunt. Pp.288. (Blackie/Halsted: 1980.) Hbk £17.95; pbk £7.95, $21.95.
Introduction to Biological Membranes. By M. K. Jain and R. C. Wagner. Pp.382. (Wiley-Interscience: 1980.) £18.40, $43.25.

THE ways in which a scientific consensus is reached on a subject must always be fascinating to philosophers of science. The consensus view on biomembrane structure is often expressed in terms of the fluid-mosaic model. At the same time it is appreciated by all the workers in the field that biomembranes exist which are not fluid but rigid, where the proteins are fixed in position, not mobile, and where a mosaic structure does not occur — instead, the proteins are arranged in a crystalline manner.

These topics and the relationship between structure and function are discussed in these two books. Both are up to date and deal with broadly the same area, covering electron microscopy, membrane preparations, structural organization, transport and receptor functions. The book by Jain and Wagner is perhaps the more elegantly written, and contains good diagrams with clear summaries and useful references, but both books will be useful to graduate and undergraduate students interested in having a concise review of the present situation.

Given two such similar books, it was interesting to see how they discussed an area of research which has excited interest over the last two or three years. This is the topic of protein–lipid interactions in biomembranes. Jain and Wagner comment

> . . . various lipid activated enzymes contain an annulus of tightly bound lipid molecules . . . . Such lipid has distinctive properties, cannot be readily extracted, is *not* readily exchangeable with the more fluid lipid . . . .

Harrison and Lunt comment

> . . . a number of membrane-bound enzymes have been shown to be surrounded by an annulus of immobilized boundary lipid, the local phase transitions of which are reflected in the activity of the enzyme . . . and boundary lipid does not preclude *rapid exchange* with surrounding lipid molecules.

Perhaps in the next editions a new consensus view on boundary lipids will be reached! However these quotations merely show how difficult it is to give a snapshot impression of a scientific area which is still alive and active. □

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Introductory chemistry in the market place

Arden P. Zipp

Fundamentals of Chemistry. 4th Edn. By F. Brescia, J. Arents, H. Meislich and A. Turk. Pp.781. (Academic: 1980.) $21.95, £13. *Foundations of College Chemistry.* 3rd Edn. By D. B. Murphy and V. Rousseau. Pp. 767. (Wiley: 1980.) $29.20, £12.45. *Basic Chemistry: General, Organic, Biological.* By D.M. Callewaert and J. Genyea. Pp.837 (Worth: 1980.) $19.95, £9.25.

THE teaching of introductory chemistry has given rise to much discussion in recent years due to two main factors. The first of these concerns the question of the topics to be taught and the level of presentation to be attempted. The second has to do with the wide variation in the abilities of the students who take such courses. These range from individuals who have received good preparation in secondary school and have little difficulty dealing with the abstract concepts which are inherent in the subject matter, to students who are ill-prepared to deal with these concepts. Although the second factor may be more common to US colleges and universities, it would be surprising if it were totally absent from classes in other countries as well.

In order to compete in the current market, textbook authors are called upon to provide books which are both broad in scope and flexible in level. The three discussed here are intended to accommodate students of varying backgrounds with potentially different career goals who need a general chemistry course; that is, students who are likely to major in science although not necessarily in chemistry. Despite this common objective, and a number of similarities among them, these books actually provide a wide choice in level and approach, and each offers something unique.

Each book is about 800 pages in length, has been produced in an attractive manner with an extensive use of colour, and all seem to have been well-edited with few errors. All three cover the topics usually treated in general chemistry: stoichiometry, atomic structure, bonding, states of matter, thermodynamics, equilibrium, kinetics, acids and bases, electrochemistry, elementary organic chemistry, polymers and (except for Callewaert and Genyea) coordination and nuclear chemistry.

Of the three, the book by Brescia *et al.* seems to be the most student-orientated, each chapter containing learning objectives, key terms, self-tests, worked-out examples and many practice problems, with answers given in an appendix. (A companion student guide is also available for this text.) Each of the other books possesses only some of these features. The authors have adopted the technique of presenting topics more than once, first on a qualitative level, then on a more intensive basis. Although this approach is successful in most cases, it is surprising to find the second stage of bonding deferred to Chapter 20 after the initial treatment has been presented in Chapter 9. The bulk of thermodynamics is presented as it relates to phase changes, rather than in a more abstract fashion. Descriptive chemistry appears mostly as a means of illustrating principles rather than in a separate chapter or chapters. A chapter dealing with environmental chemistry is presented, as is one on radiation and matter. The latter provides coverage of infrared, ultraviolet, nuclear magnetic resonance and mass spectroscopy in much greater depth than is usually encountered at this level, although some instructors may find it appropriate.

The text by Murphy and Rousseau is the highest in level mathematically and provides the greatest emphasis on physical chemistry. It presents numerous derivations including the Bohr radius, van't Hoff and Balmer equations, and the gas laws (from kinetic theory). Three centre bonds (boron hydrides) are discussed in some detail and the Debye–Huckel theory is mentioned. It is the only one of the three books in which descriptive chemistry is discussed separately. A 35-page chapter on group IV (emphasizing the variations in properties observed as the family is descended) precedes a chapter on organic chemistry, which is followed in turn by one of 57 pages in which the remaining non-metals are discussed. A separate chapter on metallurgy is also presented. The book offers many problems, both worked-out in the text and at the ends of chapters (with answers to odd-numbered ones given in an appendix), and provides a list of suggestions for further reading after each chapter. In addition, a student guide and solutions manual are available.

The distinguishing feature of the Callewaert and Genyea text is the heavy emphasis on organic and biological chemistry. Approximately one-half of the book is devoted to "general" chemistry with the remainder being divided about evenly into three sections devoted to organic molecules, biological molecules and metabolism. In addition, many of the examples used in the first portion of the text are biochemical in nature. This text was the hardest one for me to evaluate, having had little experience teaching such a course. Although the authors say that their intent is "explaining essential chemical concepts rather than providing a cursory treatment of a much larger number of topics", some of the discussions seem too brief to provide a real understanding of the material. For example, kinetic molecular theory, solids, liquids, gases and elementary thermodynamics are discussed in a chapter of 27 pages. Similarly, equilibrium and kinetics (including the disproven direct collision mechanism for the H_2–I_2 reaction) are "covered" in 21 pages. Further, it is surprising to find electron configurations relegated to an optional section of a chapter and to encounter a discussion of bonding without substantial use of orbitals. This approach may be appropriate and even necessary in order to cover all the material included in the text (and presumably in such courses), although the book's use in the broad general chemistry market is likely to be limited. The book does give study objectives, useful chapter summaries and solutions to exercises at the end of each chapter, but has relatively few problems and worked-out examples and is the least mathematical of the three.

These three texts represent only a small sample of the total number which is currently available, although they do provide some indication of the almost limitless variety which exists in this highly competitive market. □

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Biophysical challenge

R. G. Gosling

Topics in Classical Biophysics. By H. J. Metcalf. Pp.286. (Prentice-Hall: 1980.) Pbk £6.45, $9.95.

MY readiness to review this book lay in its intriguing title and my need of a broadly based text for students pursuing a BSc degree in "physics with medical applications". The format is primarily that of a textbook, although the author suggests it "is not intended for students alone". The first eight of its nine chapters present appropriate basic theory — biomechanics, heat and energy, fluids, blood circulation, feedback and control, nerve cells, sound and hearing, light, colour and vision. The final chapter is a somewhat mixed bag entitled "Experimental Techniques" but, with its emphasis firmly on the use of physics to acquire information, nicely draws together the joint application of various principles previously discussed.

Essentially, the theoretical treatment is that of classical continuum physics, with basic equations developed at the beginning of each chapter and then applied to some selected topics appropriate to that field. Whilst the question as to whether this justifies the title *Classical Biophysics* could be debated, I found that the material chosen represented a useful introduction for any undergraduate interested in the applications of physics to medicine. The only pre-requirement is an elementary

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knowledge of calculus, for example additional maths at "O" level in the UK or an introductory year college math course in the USA.

Because of the wide range of natural phenomena discussed, the theory that can be developed in the commendably brief compass of 275 pages is necessarily kept at a lowish level. The penalty is that arguments are sometimes terminated a little prematurely. For example, in Chapter 3 on fluids, the forces acting in a Newtonian liquid flowing in a rigid tube are clearly deduced, but then used only in the special case of a constant pressure gradient. This precludes any application of such theory in discussing arterial haemodynamics in the next chapter. However, it does provide a clear "launching point" for any lecturer using the book as a class text.

It is my impression that this book will present first- and second-year students of physics, biology or medicine with challenging reading and I look forward to using it with such classes next year. It may prove a useful introduction to any courses concerned with the application of classical physics to biomedical problems. □

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Food — science and politics

J.W.T. Dickerson

Nutrition: in Perspective. By Patricia A. Kreutler. Pp.700. (Prentice-Hall: 1980.) £12.30, $18.95. Workbook also available.

THERE has recently been something of a spate of books on nutrition, and new ones must therefore face increased competition. The market itself has also increased, for we are becoming much more aware of the relevance of food to life and health in the modern world. Moreover, all shades of interest and competence, from the lay reader to the academic, can profit from the study of the subject. This book has been aimed somewhere in the middle of the range. It "has been written to provide students with an understanding of the science of nutrition"; to acquaint them "with the issues facing nutritionists, scientific and government leaders, and consumers in contemporary society", and "to enable students to translate knowledge into practice".

The first part of the book is concerned with the scientific principles of nutrition, and contains nine chapters. The second part discusses aspects of nutrition for everyday living, in seven chapters. The author and publishers are to be congratulated on the plan and layout of the book. The text is clear, direct and liberally illustrated throughout with tables, figures, photographs and the occasional cartoon. The author is clearly an expert communicator of her subject. References in the text have been kept to a minimum and are listed, with recommendations for additional reading, at the end of each chapter.

A feature of the book is the introduction, as appropriate, of "Perspectives" — applications to health or topics of contemporary concern. Thus, in the chapter on carbohydrates, the perspectives are sugars in foods, diabetes and hypoglycaemia, and dietary fibre. The chapter on lipids contains a perspective on diet and heart disease. Perspectives on sodium and hypertension, iron enrichment and the politics of fluoridation are inserted in the chapter on minerals, whereas fad diets are discussed in the chapter on food in contemporary society.

The whole book is a pleasure to read. It is interesting, informative and contains a great deal of valuable information. □

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Chemistry for life

Peter Garratt

Essential Organic Chemistry for Students of the Life Sciences. By A.P. Ryles, K. Smith and R.S. Ward. Pp.306. (Wiley: 1980.) Hbk £14.90, $44.10; pbk £5.25, $15.75. *Organic Chemistry: The Basis of Life.* By Bernard Miller. Pp.472. (Benjamin/Cummings/Addison-Wesley: 1980.) $18.95, £11.85.

THE problem of teaching chemistry to students in the biological sciences is one of striking a balance between relevance and comprehension. The student, who probably has no great interest or aptitude for chemistry, wants only that material which is immediately relevant to his biological studies, whereas the teacher knows that to have any appreciation of the role of chemistry in biology the student must understand the concepts and principles on which the subject is based. These two books, although emanating from different continents and very different educational systems, clearly illustrate that this problem is also felt by writers of texts for such students.

Both books deal, as their titles imply, only with the organic component of the chemistry required by biologists. RSW (for short) has three introductory chapters, the first on bonding, functional groups and mechanism, the second on stereochemistry and the third on techniques. Then follow nine chapters presenting a conventional and systematic treatment of organic chemistry from the alkanes to the proteins and nucleic acids. The last three chapters deal with tetrapyrroles, physiologically important compounds and metabolism and biosynthesis. Miller has an introductory chapter in which "basic principles" are sketchily and sometimes erroneously treated, followed by a chapter on the alkanes whch includes further introductory material on bonding. The remaining 16 chapters give a similar outline of organic chemistry to that found in RSW, except that the ordering of material differs. For example, chirality is introduced on p.23 in RSW but not until p.197 in Miller, rather late for a book subtitled the *Basis of Life*. Miller integrates the biological examples more intimately with the chemistry than do the British authors, and each chapter ends with a summary of the important principles and concepts found therein. Both texts have problems distributed within the chapters, and there is a student study guide available for Miller and one promised for RSW.

The layout, appearance and style of a text are important to its student user. Both texts are well produced, Miller being a two-colour printing, in which the colour has been judiciously used, while RSW is in monochrome. Miller has clear structural diagrams and well drawn, and therefore useful, representations of space-filling models, but it also has unnecessary photographs of oil rigs and similar irrelevant objects. RSW has, in general, clear structural formulae except for those representing the conformations of cyclohexane and for ascorbic acid, the nicotinamide co-enzymes and co-enzyme A. RSW also uses Haworth rather than conformational formulae for the carbohydrates. Since shape is so important for biological activity, conformational structures are much more instructive, and the treatment in Miller, which uses such structures, is clearly superior.

Both of these texts would form a satisfactory basis for a course in organic chemistry for biologists, but whether much would be gained over using a standard chemistry text such as Solomons' *Organic Chemistry* (Wiley, 2nd Edn 1980 — see over for review) is a moot point. Miller certainly makes a more direct attempt to show the relevance of the chemistry to biological molecules and processes than does RSW and the style is easier to read, but the sections on orbitals and bonding would require remedial attention from the lecturer. □

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Organic chemistry, in theory . . .

John Mann

Organic Chemistry. 2nd Edn. By T. W. G. Solomons. Pp.1066. (Wiley: 1980.) Hbk £15.25, $33.25; pbk £7.90, $18. *Organic Chemistry*. By D. S. Kemp and F. Vellaccio. Pp.1421. (Worth: 1980.) Hbk $26.95, £10.25; pbk $14.95, £5.

IT IS not often that an organic chemistry textbook is the subject of a plagiarism suit, but such was the fate that befell the first edition of *Organic Chemistry* by T. W. G. Solomons. After a four-month hearing the book and its author were cleared in March 1980, just prior to the release of the second edition. Personally, I have always found the other book in question (*Organic Chemistry* by R. T. Morrison and R. N. Boyd; Allyn & Bacon, 3rd Edn 1973) pretty indigestible and rather old-fashioned, though students seem to like the summaries of functional group chemistry.

Professor Solomons's book has similar summaries at the end of most chapters, but it also has a great many features which enhance its value relative to Morrison and Boyd. The order of treatment is conventional, commencing with hydrocarbons and bonding, and proceeding through the various functional groups to carbohydrates and proteins. Well-chosen special topics are interspersed throughout, and complement the chapters in which they appear. We thus have naturally-occurring alkenes and the photochemistry of vision at the end of the chapter on conjugated unsaturated systems; a beautifully illustrated and lucid account of protein biosynthesis accompanies the chapter on amino acids and proteins; and there are also useful sections on applications of the Woodward–Hoffmann rules, halo-organic insecticides, polymerization, and biosynthesis of fatty acids and steroids. One small puzzle is why thallation deserves a six-page section of its own, while the Wittig reaction and organosulphur chemistry (both much more important in modern organic chemistry) receive only scant attention. The chapter on spectroscopic techniques, now a must for any organic text, provides a very good introduction to the subject, and is comparable with the corresponding section in *Introduction to Organic Chemistry* by A. Streitweiser and C. H. Heathcock (Macmillan, New York, 1976), the book that we recommend to our students.

There are a few disappointments in Professor Solomons's book. For example the absence of a section on the strategy of synthesis, and the rather old-fashioned account of synthesis and reactions of β-dicarbonyl compounds which contains a long list of named reactions, but little in the way of modern synthetic applications or mention of their relevance to biochemistry. These criticisms aside, the main functional group chemistry is well done, the book is beautifully produced with two-tone drawings and section headings, and there are clear chemical structures throughout. It is readable and reasonably up-to-date in most areas, and has innumerable useful problems throughout, with answers supplied at the end of the book or in the separate solutions manual.

The authors of the second glossy American book under review, D. S. Kemp and F. Vellaccio, claim that there is a need for a text emphasizing the chemistry of the oxygen- and nitrogen-containing functional groups. The book is designed for an American one-year/three-semester course (as I suspect Solomons's book is), and it is divided into three main parts: chemistry of functional groups and structure proof; chemistry of hydrocarbons and carbonyl chemistry; and advanced topics. The order of treatment is thus somewhat different from that of most other texts. Apart from this difference, the treatment of topics is rather similar to that in Solomons, except that there is more emphasis of applications to organic synthesis. The authors claim that their treatment of carbonyl chemistry is the "most accessible and interesting of any that are available", and in this they are probably correct. The chapter on spectroscopic methods provides a good introduction to NMR and IR, but is very weak on mass spectrometry (a mere three and a half pages), while chapters on carbohydrates, proteins and nucleic acids are good, though not so well done as in Solomons. There are good sections on the Woodward–Hoffmann rules; rearrangement reactions; reactive intermediates; and organometallic reagents, which includes as special topics hydroboration, silanes, organomercury compounds and thallation. Coverage of heterocyclic chemistry is, however, rather weak, and natural products are dealt with in a somewhat fragmented way. The penultimate chapter, on the synthesis of six compounds of theoretical or practical importance, is novel and makes interesting reading. It includes, amongst other topics the conversion of penicillins into cephalosporins, and capped porphyrins. Again, there are useful problems throughout, and the 670-page workbook and solutions manual should find many friends amongst both students and academics. Throughout, the illustrations are very good, but the book is not quite so lucid or readable as Solomons, and many students will miss the reaction summaries.

Both books are modern texts comparable to the excellent Streitweiser and Heathcock, and will I am sure sell well, especially in the USA. However neither of them has everything quite right, and the ultimate text has still to be written. But then, there is probably no such thing! □

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. . . and in practice

Paul Schatz

Experimental Organic Chemistry. By Michael P. Doyle and William S. Mungall. Pp.490. (Wiley: 1980.) Hbk £10.10, $23.70; pbk £5.95, $13. *The Systematic Identification of Organic Compounds*. 6th Edn. By R.L. Shriner *et al.* Pp.604. (Wiley: 1980.) Hbk £13, $30.55; pbk £6.90, $15.90.

THE pedagogical aim of an organic laboratory course is to introduce the student to the basic techniques used by chemists for the preparation, separation, purification and identification of organic compounds. Besides demonstrating such techniques as distillation, recrystallization, chromatography and extraction, the laboratory is a showplace for demonstrating the chemical properties of the various organic functional groups. Most introductory laboratory courses use what might be dubbed "the synthetic approach". The techniques are introduced in the context of carrying out typical organic reactions, such as an Sn1 substitution or an E2 elimination, isolating and purifying the product, and confirming the identity of the product. An alternative approach to teaching in the organic chemistry laboratory could be called "the characterization approach". In this approach, the laboratory techniques are taught in the context of classifying and identifying organic compounds by functional groups. Typically, derivatives of compounds are prepared and purified, and then identified by measuring physical properties. Although most qualitative analysis courses are usually reserved for intermediate or advanced level undergraduate students, many modern introductory courses employing the "synthetic approach" also incorporate a qualitative analysis component into the curriculum.

There are a large number of textbooks (or laboratory manuals) available for the introductory laboratory and the manner in which they treat the material is very similar. There is a section introducing basic techniques, there is a section on qualitative analysis and there is a section containing a variety of experiments for the instructor (or

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student) to choose from. *Experimental Organic Chemistry* follows a similar pattern. What distinguishes it from other texts are the experiments. They are not a rehash of familiar experiments, but a set of new ones. Moreover, they are interrelated since the product of one experiment can serve as the starting material for another. The authors suggest that besides being educationally beneficial, it can also whittle down the cost of the laboratory course. By choosing readily available chemicals for both the starting materials and the products, the authors have avoided the pitfall of locking the user into a prescribed set of experiments and the vexing problem of what to do with the student who gets a poor yield. The variety of experiments is impressive — Sn1 substitution, Sn2 substitution, enzymatic hydrolysis, phase transfer catalysis and so on — and they include a good collection of short multistep (two or three steps) syntheses. The book has much to recommend it besides the experiments. The techniques are explained fully and clearly, the experimental procedures are detailed and clear, the artwork is accurate and well drawn, and the reproductions of the IR and NMR spectra are sharp. All in all, this is a well crafted book, definitely one to be considered when choosing a text for laboratory courses.

One of the most familiar books for the organic qualitative analysis course has been *The Systematic Identification of Organic Compounds.* A sixth edition has been recently published and this edition is markedly different from its predecessors. The most obvious difference is the size; the newest edition is approximately 50% wider than the fifth edition. This larger size is needed to contain the expanded text. For the most part, all of the elements of the older editions are incorporated in this edition, including the measurement of physical properties, the classification tests, the preparation of derivatives and the comprehensive tables of derivatives. What has been added are more extensive discussions of spectroscopic methods of identification and chromatographic methods of separation. The latter topic was completely omitted in previous editions.

Other additions are a larger introduction, including a discussion of laboratory safety, a more detailed explanation of basic laboratory techniques such as distillation and recrystallization, and a chapter on the literature of organic chemistry (principally an annotated bibliography). The book would be useful for the classic organic qualitative analysis course (if any of these still exist). It could also be used in an introductory organic laboratory course emphasizing qualitative analysis if supplementary material describing the theory of the basic techniques is available, since no theory is explained in the text. □

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Joys of general chemistry

Charles A. McAuliffe

Chemistry (With Inorganic Qualitative Analysis). By T. Moeller *et al.* Pp.1085. (Academic: 1980.) £14, $21.95. *Chemistry: A Systematic Approach.* By Harry H. Sisler, Richard D. Dresdner and William T. Mooney Jr. Pp.960. (Oxford University Press: 1980.) £13.50, $19.95. *First Year Chemistry.* By J. M. Coxon, J. E. Fergusson and L. F. Phillips. Pp.376. (Edward Arnold: 1980.) Pbk £9.75.

I HAVE been reviewing freshman chemistry texts for this edition of *Nature* for four years and, despite a long association with a number of American universities and their methods of teaching, each particular year of carrying out this task offers me an opportunity of seeing new facets in these books. For example, the best of this type of book, in addition to expounding chemical fact and theory, also stresses the application of chemistry to everyday life. I have been particularly pleased by attempts in a number of excellent texts at the way in which environmental problems are related to the chemistry being illustrated. For example, consideration of the reactions involved in the generation of photochemical smog cannot only make a student appreciate the chemistry of the nitrogen oxides and ozone, but also that of organic free radicals and the importance of light in initiating chemical reactions. Similarly, dealing with elemental cycles in the biosphere not only emphasizes their importance to life but also serves to make simple chemistry very real; for example, the most important of all chemical processes, photosynthesis, should not be dealt with in isolation, large subject though it is, but can in a freshman text be linked to the oxygen and carbon cycles and fossil fuel generation.

The enormous market for freshman (or general chemistry) texts in the United States ensures that many are available. In previous years I have emphasized how awful some of these texts are, and one shudders to think of them being foisted on students by their authors; many potentially creative chemists must be lost in this way. However, the three texts dealt with here are, in their different ways, a joy to behold.

Chemistry (with Inorganic Qualitative Analysis) has so much to recommend it. There are 32 chapters and it is sensible to give a brief indication of their range so that one can understand the scope of a large textbook such as this. Atomic structure and nuclear chemistry, chemical bonding, periodic properties, thermodynamics, kinetics and electrochemistry provide an adequate theoretical basis. Descriptive group chemistry and some, though never enough, organic chemistry is related to reactivity and stoichiometry. There are several chapters on qualitative analysis.

It is difficult to single out individual sections of this book for praise. I used the extensive index a great deal in examining the book. Nothing which should be included has been omitted; the text is concise and illuminating. Like many such books, the diagrams are excellent. I particularly enjoyed the copious vignettes entitled "Thoughts on Chemistry". These are enlightening quotes, a page or so in length, which range from "The Requisites of a Good Hypothesis" (Robert Boyle), "The World's Biggest Membrane" (from Lewis Thomas's memorable observations in *The Lives of a Cell: Notes of a Biology Watcher*), "The Chemical History of a Candle" (Michael Faraday), "The Centrality of Chemistry" (Martin Sherwood), to "The Epigrams of Remigius Fresenious". This book is to be highly recommended. I have only two criticisms. The minor criticism is that on the same page (p. 21) which shows the structure of chlorophyll *a* there is advice on how to pronounce $NaNO_3$ ("N-A-N-oh-three") and H_2SO_4 ("H-two-S-oh-four") — this grated on me. More serious is the large amount of space they devote to inorganic qualitative analysis. This is a grindingly boring topic and, if included at all, should be kept to a minimum. The authors would do well to extend their good descriptions of infrared, ultraviolet and nuclear magnetic resonance spectroscopy.

Chemistry: A Systematic Approach is also to be recommended. It is comprehensive and I particularly enjoyed the chapter on the chemistry of living systems. An instructor's manual (by W. H. Myers) is available.

First Year Chemistry is a somewhat different book. The authors aim to produce a text in which approximately equal weight is given to physical, inorganic and organic chemistry, and minimizes the divisions. It is a very good attempt, but the book may have limited appeal. It assumes a much greater basic knowledge than do the other texts and may be only suitable for the New Zealand system, in which the authors work.

Finally, might I appeal to all of these authors? Expressing photosynthesis as

$$6CO_2 + 6H_2O \rightarrow C_6H_{12}O_6 + 6O_2 + \text{energy}$$

is very misleading, even though this is the way major biology texts frequently express it. Carbon dioxide does not react with water as implied by the equation. Further explanation would be enlightening □

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Quantum mechanics for chemists

A. J. Stone

Orbitals and Symmetry. By D. S. Urch. Pp.256. (Macmillan: London, 1979.) Pbk £5.95. *Chemical Structure and Bonding.* By R. L. DeKock and H. B. Gray. Pp.491., (Benjamin/Cummings/Addison-Wesley: 1980.) $21.95, £9.95.

THE flood of textbooks on quantum mechanics for chemists puts one in mind of that wilderness of monkeys which are supposed in time to produce the works of Shakespeare — not because the results are similar but because we are prompted to ask: why bother? We already have textbooks at every level which give clear, sound coverage of the principles of the subject. It is not enough for a new book to do the same; it must provide new insights, take account of advances in experimental and theoretical technique, or treat some aspect of the subject substantially better than before.

DeKock and Gray's angle is to relate the discussion of bonding and structure to the results of photoelectron spectroscopy. However, the book begins with the Bohr theory (what other superannuated theory has such resilience that it is still being taught 50 years after it was superseded?) and there is much emphasis on Lewis octets and valence-shell–electron-pair repulsions. Valence-bond theory is discussed at length, and molecular-orbital theory takes a bad second place. For example, a superficial and rather inaccurate account of a simple Walsh diagram is followed by the statement that "predicting molecular shape using molecular-orbital theory . . . is much more tedious than the valence-shell electron-pair repulsion method". This attitude to molecular orbital theory is curious in view of the detailed account of the relationship between photoelectron spectroscopy and bonding, which is the one useful feature of the book and which is done rather well — indeed, at a level which is noticeably higher than the rest of the book.

Urch's book is a reprint of a volume originally published in 1970 in the abortive Penguin Library of Physical Sciences, and although the preface implies some revision of the text, this version looks virtually identical to the original. Some errors have been corrected, but others have not, and the bibliography remains unchanged. My main criticism is that there is not enough attention given to the business of showing the student how to do the calculations. For example, the representation matrices for a set of d orbitals in the octahedral group (a notorious source of difficulty) are simply written down without comment. More importantly, the procedure for obtaining symmetry orbitals (the projection formula) is never mentioned, and symmetry orbitals are blandly produced, not merely without explanation but without any apparent appreciation that the reader might not see where they came from. The text could quite easily have been revised to correct these failings, but the opportunity has been lost and a potentially useful account remains incomplete. □

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Environmental chemistry, theory and practice

Roy M. Harrison

Environmental Chemistry: The Earth–Air–Water Factory. By R. W. Raiswell *et al.* Pp.184. (Edward Arnold/Halsted: 1980.) Flexi £4.95, $14.95. *Experiments in Environmental Chemistry: A Laboratory Manual.* By P. D. Vowles and D. W. Connell. Pp.102. (Pergamon: 1980.) Hbk £10, $23; pbk £4.50, $9.95.

THE teaching of environmental chemistry as a component of environmental science, ecology and chemistry courses at university level has developed and expanded considerably over recent years, largely in the absence of adequate textbooks. A number of books have appeared recently, but there still exists a need for sound teaching texts.

Environmental Chemistry: The Earth–Air–Water Factory provides a broad introduction to the field written mainly from the viewpoint of the geochemist. The factory analogy is used apparently in an attempt to provide a better integration of subject matter and to encourage the student's appreciation of the strong links between the various areas of environmental chemistry. The book assumes rather little chemical background and even includes a glossary of chemical terms. It is written with clarity by authors acknowledged as authorities in their respective fields. The great detraction, however, is that no text of 154 pages (excluding glossary and index) can more than touch the surface of this vast subject area. It is difficult to see the book being used for university teaching other than at the most basic level as it will satisfy neither the lecturer's aims nor the student's curiosity.

Experiments in Environmental Chemistry: A Laboratory Manual concentrates upon pollution and ecological chemistry, the majority of experiments being analytical in nature. The presentation is similar to that used in laboratory manuals in universities, with an introductory section followed by experimental instructions and then guidance for writing up. The factual material is occasionally unreliable, although in some instances this arises from typographical errors. The experiments require a fair variety of analytical equipment which may be beyond the resources of some teaching laboratories, and some demand rather problematic starting materials — human milk; butterfly larvae and plants from the Southern Hemisphere; replicate 0.2 g samples of household paint removed from surfaces, for example. The book contains some excellent ideas for experiments but is more likely to find use as inspiration for the teacher than as a laboratory manual used directly by the student. □

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Environmental what?

John Barkham

Ecology and Environmental Management. By C. C. Park. Pp. 272. (Butterworths/Westview, Boulder, Colorado: 1980.) £12.50, $26.25. *Natural Resource Conservation: An Ecological Approach.* 3rd Edn. By Oliver S. Owen. Pp.883. (Macmillan, New York/Collier Macmillan, London: 1980.) £12.95, $19.95. *Introduction to Environmental Science.* By J.M. Moran, M.D. Morgan and J.H. Wiersma. Pp.658. (W.H. Freeman: 1980.) £19.95, £9.80. Instructor's guide also available.

MANY different disciplines contribute to attempts to understand the environment. The rapid development of courses in environmental sciences or studies in recent years reflects the need to look beyond geography, geology, biology or economics alone for the range of material required. However, the variety of university and college courses calling themselves "environmental" suggests widely differing views as to what are the key subject areas, concepts and techniques to which students should be addressing themselves. That there are degree courses in "environmental *science*" implies that there are those bold or unwise enough to define a distinct discipline. A more sensible and pragmatic approach is to regard the sciences concerned with the environment as a federation. They may usefully be gathered together in one department so that environmental problems may be explored from a

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number of different perspectives. This flexible view allows students to adopt an eclectic approach to learning, drawing upon sources of information which appear to be useful whatever their pedigree may be. So it is likely that books written about environmental sciences from a variety of points of view will be useful to different students at different times.

The three books considered here all examine problems of environmental management from an ecological perspective. They appear to be directed towards first-level university and college courses. Questions of management always stimulate student interest and passion, but the soundness with which undergraduates approach such issues usually depends upon the adequacy of their grounding in basic biological, environmental and social sciences. All three books start from the assumption that their readers know little or no ecology and attempt to give them a grounding in it. Park's book, *Ecology and Environmental Management,* devotes about 170 pages to this material, about two-thirds of the book, although the first 20 pages are spent in largely sterile justification of the notion that geography is "centrally placed to contribute effectively to the solution of many environmental problems". This section rather underlines the fact that this is an ecological text written by a geographer for geographers and environmental scientists. Ecologists will find the balance rather odd and it is by no means clear at times why, in relation to environmental management, we should be learning the details of some ecological processes. There are some extraordinary statements, too, including a comment on the ". . . lack of interest within ecology for animal populations . . ." (p.39). For me, there is a definitely dated feel to the first two-thirds of this book, which is perhaps emphasized by the predominance of geographical references and the paucity of ecological ones from the 1970s.

In Owen's book, *Natural Resource Conservation*, the ecological basis provided is the barest minimum necessary and is rather scattered about within the text following a short section near the beginning. Moran, Morgan and Wiersma's *Introduction to Environmental Science* is the best balanced from this point of view, although the title is entirely misleading. Ecological fundamentals occupy about as much space as in Park's work, the approach is traditional — but none the worse for that — and, above all, the material is well organized. It is obviously a carefully planned book and I wonder whether this is the pay-off for good teamwork. There are conclusions, useful summary statements, review questions and boxes of information where deeper explanation of a topic is felt necessary.

In the environmental management sections, Owen's and Moran *et al.'s* American texts are very different from Park's English approach. Having waded through Owen I felt blasted with the sheer volume of material on every kind of resource and pollution problem. No question of a balanced view here. I felt the author wanted the reader to join him in the corral of the good boys. However, there is a lot of qualitative information about every kind of problem, solutions are discussed and the work is liberally illustrated. Moran *et al.* cover the same ground in about the same amount of space but in a well-ordered, less frenetic manner and with fewer gee-whiz statistics. Both books use very largely American case studies and examples and refer to American ways and means of tackling these problems.

Park's book examines resource, pollution and management issues relevant to Britain. Only two pages are devoted to problems of a global scale. Students concerned with nature conservation will find a lot of useful material, particularly in the tables. Others taking a broad-based course will get a reasonable overview of the subject.

In terms of a best buy, Owen's book may well appeal to a section of the American market, but not in Britain. Park's will have a limited appeal mainly to geographers in Britain, but not in America. Moran *et al.* is best value for money and may sell to a limited extent in Britain as well as in America, although a title such as *An Ecological Approach to Environmental Management* would be more appropriate. Sadly, none of these books succeed in giving the student a clear global perspective which is not at the same time banal. It is also clear to me that any student who is exposed only to the approach adopted here will make a disastrous environmental manager. □

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Views of the landscape

C. Kidson

Process in Geomorphology. Edited by C. Embleton and J. Thornes. Pp.436. (Edward Arnold/Halsted: 1979.) Hbk £16, $44.95; pbk £8.95, $19.95. *Geomorphological Processes.* By E. Derbyshire, K. J. Gregory and J.R. Hails. Pp.312. (Butterworths/Westview, Boulder, Colorado: 1979.) Hbk £12.50, $31.25; pbk $13.50.

IT IS now more than 30 years since the famous symposium, commemorating the centenary of the birth of W. M. Davis, marked the beginning of the so-called "new" geomorphology of which these two texts, in their differing ways, are examples. In 1952, in "The Dynamic Basis of Geomorphology" *(Bull. geol. Soc. Am.* **63**, 923–938) Strahler amplified the "dynamic approach" advocated at the symposium, which required the quantitative determination of landform characteristics and the formulation of empirical equations by the methods of mathematical statistics. This "counterbalance" which, he felt, should replace the Davisian explanatory descriptive method, involved

> the study of gravitational and molecular shear stresses acting on the materials of the earth's surface, which behave as elastic or plastic solids, or viscous fluids to produce the characteristic varieties of strain or failure that constitute weathering, erosion, transportation or deposition.

Process in Geomorphology and *Geomorphological Processes* both adopt this approach but with differing degrees of emphasis and with differing success. The first is almost a caricature where the landform characteristics appear frequently to be forgotten and mathematical statistics and empirical equations seem, at times, to be incorporated for their own sakes rather than to make an understanding of the operation of processes easier. Indeed, some of the authors have succeeded in making relatively simple topics seem difficult. Even final-year undergraduates will have difficulty with parts of this volume, especially if they have no background in physics and hydraulics. The excellent introductory chapters go some way towards helping such students. *Geomorphological Processes* succeeds in adopting a rigorous approach but in a more balanced way, maintaining a clarity of mathematical as well as of verbal expression which illuminates rather than confuses.

In *Process in Geomorphology* two other defects mar what is, nonetheless, a useful compendium of scholarly essays. The idea that landforms may encapsulate the results of many processes, of which those now seen in operation are but the most recent, finds no place in the book. Changes of climate and of process over time, for example those resulting from the advances and retreats of Pleistocene ice sheets, are largely ignored. Second, rates of operation of processes, particularly in the context of the rare event, are inadequately considered by most of the authors. The concluding chapter by Dr Thornes does nothing to remedy these defects.

Geomorphological Processes similarly concentrates on contemporary processes and to some degree suffers from the same defects. However, the short introductory chapter makes clear that this is a conscious choice and indicates the need to examine these other facets of landform developments, at greater length, elsewhere.

Derbyshire, Gregory and Hails succeed in making their study an exercise in

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of the landscape, by constantly maintaining the link between process and form. This is made possible by the arrangement of the book. Essentially, processes are divided into glacial and periglacial, fluvial and marine with only a small chapter devoted to the work of the wind. A much shorter introduction than in the Embleton and Thornes volume allows each of the three major sections to be treated at greater length. The effect is most marked in Hails's chapter which is a full and authoritative treatment of coastal processes. By contrast, Clark in *Process in Geomorphology,* asked to attempt a similar geomorphology, rather than in the physics exercise in half the space, not unnaturally makes a more superficial presentation.

These two volumes are interesting for contrasts in style rather than in approach. Both have value. *Process in Geomorphology* will be useful to the readers to whom it is directed much more as a reference work than as a course primer; *Geomorphological Processes* could well satisfy the opposite function. □

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Sedimentary, my dear Watson!

Bruce Sellwood

Origin of Sedimentary Rocks. 2nd Edn. By H. Blatt, G. Middleton and R. Murray. Pp.782. (Prentice-Hall: 1980.) £20.10, $30.95.

UNTIL the middle of this century, the study of sedimentary rocks fell largely under the two headings of stratigraphy and petrology. Sedimentology as a separate integrative discipline emerged no more than 30 years ago and a major surge in the subject has occurred over the past 20 years, much of the impetus for new developments coming from the demands of the oil and mineral extraction industries.

One of the first integrative modern sedimentological texts was *Origin of Sedimentary Rocks* (Prentice-Hall, 1972) which stressed the genesis of sediments and the processes of physical and chemical sedimentation. At that time the book was generally welcomed as a well organized, comprehensive and admirably clear text. After eight years, a totally revised edition has appeared. The new edition is almost 25% longer than the original but retains the successful structure and stylistic clarity of its predecessor. The book is divided into six parts and contains 19 chapters, each of which ends with a useful annotated reference list.

Part I introduces the scope of sedimentology, emphasizing the role of field investigation. Discussion of various facets of the geological cycle is undertaken in Part II.

Part III is a review of terrigenous clastic sediments, and commences with a new and welcome chapter on the geochemistry of natural waters. This is followed by coverage of weathering processes and products, mineralogy of sandstones, cementation and sandstone diagenesis, and sandstone classification. The chapter on cementation and diagenesis is also new and reflects the recent developments in this field which stem directly from petroleum reservoir studies.

Part IV considers the origin, diagenesis and classification of limestones, dolomites and evaporites, while Part V tackles the disparate topics of chert, phosphates, iron-rich rocks and manganese nodules.

The final part is devoted to facies models. Although the nucleus of this section appeared in the first edition, it has been largely re-written and greatly expanded. It emphasizes the predictive nature of facies analysis and environmental interpretation and, although larger than the original version, is still briefer than it should be in view of the importance of accurate facies modelling to economic geology.

There are no attempts at facies modelling using electric-log analysis, which is a pity since these methods are so important in contemporary hydrocarbon exploration. Some diagrams in the first edition were unclear and have been redrafted, but there are still a few over-reduced diagrams or diagrams taken from poor originals. Yet these are minor quibbles.

Overall, this is an excellent general source text for students and professionals. I hope it appears in paperback — soon.

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New mineralogy

D. Flinn

Principles of Mineral Behaviour. By A. Putnis and J. D. C. McConnell. Pp.257. (Blackwell Scientific/Elsevier/North-Holland: 1980.) Hbk £18, $45; pbk £9.80, $24.95.

THIS book is the first of a new Blackwell series entitled *Geoscience Texts;* it is also the first new mineralogy textbook for undergraduates to appear for a very long time. Each year a number of ostensibly new texts appear but on examination they turn out to be updated or merely rehashed versions of books which have been with us for decades, or even since mineralogy began. Descriptive mineralogies in one guise or another are legion, and optical crystallographies seem to appear every year, all closely modelled on *Optical Crystallography* by Whalstrom which first appeared in 1943 and now is in its fifth edition (Wiley, 1979). For years I have had to refer my students, for accounts of mineral behaviour, to odd chapters in texts written for physicists, ceramicists, metallurgists and lately geochemists.

Now, at last, such accounts have been put together in one book and brought bang up to date for geology undergraduates. The book is descriptive, being concerned with kinetics rather than equilibria, but the necessary minimum of thermodynamics is clearly and simply presented. The approach is process orientated rather than mineral orientated even though the processes are all demonstrated in common minerals. The authors concentrate on polymorphism and on the sub-solidus behaviour of solid solutions and show how the end products depend more on the time available than on equilibrium relations. I hope that in future publishers will use the resources they now devote to publishing new optical crystallographies to updating this type of textbook and expanding it to cover other types of mineral behaviour such as crystal growth and crystal deformation. □

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Extended senses

Robert N. Colwell

Remote Sensing: Optics and Optical Systems. By P.N. Slater. Pp.575. (Addison-Wesley: 1980.) $34.50, £20.70.

FOR years, remote-sensing educators and their students, as well as engineers and researchers in remote-sensing-related fields, have needed a comprehensive book which would explain, in a readily understandable way, both the principles and the design of various kinds of remote-sensing instruments, and the procedures best used in measuring the performance of such instruments. Professor Slater's book seeks to satisfy that need.

In this book, of 14 chapters and 9 appendices, a systematic treatment is given of the various optical and electro-optical systems that are currently used for remote sensing in the 0.4 to 16μm range, including the Landsat multispectral scanner, return beam vidicon and thematic mapper systems. (Microwave remote sensing — both active and passive — is reserved for

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another book in the series.) In particular, the author describes in detail the appropriate methods for correcting the "spectral signatures" of features for both atmospheric and system-dependent effects. A substantial amount of the author's own high-quality research is integrated into the book; however its greatest strength lies in the clarity with which Slater has explained, using little more than basic physics and simple mathematics, the relevant work of scientists in a great variety of fields, ranging from human optics to electromagnetic theory, from electronic circuitry to spectroradiometry, and from basic atmospherics to matter/energy interactions at the Earth's surface. Of particular value is his consistent use throughout the book of a powerful performance criterion, the "modulation transfer function".

Despite the prodigiousness of his undertaking, Slater is careful, for the most part, to cite references to the many principles with which his book deals. Thus it is all the more disconcerting when, on occasion, he makes flat assertions (without providing documentation) such as "the average eye can discriminate approximately 5 million colors but only 200 shades of gray". Actually there are almost as many (widely disparate) values quoted in the literature as there are experts who have addressed this highly important matter. An occasional citation is improperly listed in Slater's book but such errors will no doubt be corrected in future editions.

All told, Slater's book is highly authoritative, unusually up to date and comprehensive, clearly written, well fortified with relevant tables and neatly drawn figures, and eminently worth perusal by any serious remote-sensing scientist. □

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Beyond Wiener

Freeman Gilbert

Geophysical Signal Analysis. By E.A. Robinson and S. Treitel. Pp.466. (Prentice-Hall: 1980.) £23.40, $36.

GEOPHYSICAL signal analysis became a subject of research by the authors in the early 1950s, when they were graduate students at MIT. They were members of the Geophysical Analysis Group, sponsored by a consortium of petroleum and exploration companies, whose rather ill-defined goal was to use digital computers for the application of the ideas, techniques and methods of Norbert Wiener to the interpretation of reflection seismograms. In the intervening quarter-century the seismic exploration industry has "gone digital" in a big way. In a typical day the industry processes several thousand computer tapes (9 track, 1600 BPI, 2400 feet) of data on multiple mainframe computer systems. By contrast, in the early 1950s the authors used the Whirlwind II computer (now in the Smithsonian Museum). Input was on punched paper tape, output on electric typewriters and secondary storage on magnetic wire; instruction time was in milliseconds and memory was 1024 16-bit words. It was the premier computer of its day. The authors deserve a large part of the credit for developing many of the currently used computational procedures in geophysical signal analysis and their book provides the basis for understanding and using them. Their firsthand knowledge and experience are evident throughout.

The method of least squares pervades science and engineering; it is the central theme of the present volume. Basically the book is concerned with filtering, the process of removing or diminishing "noise" in order to reveal and enhance the "signal". With the reflection seismogram as a model, the authors develop least squares digital filtering from first principles in the first eight chapters. The subjects of minimum phase lag, minimum delay, causality and stability are presented in a thorough manner, and the concept of a prediction filter is introduced. The writing is well balanced between theory and application. This arises from the blending of the authors' talents — Robinson is a mathematician at home with geophysics and Treitel is a geophysicist at home with mathematics. They complement one another very well.

The second half of the book is devoted primarily to the subject of deconvolution, especially predictive deconvolution. The opening chapter is devoted to a discussion of stationary time series. The method of generalized harmonic analysis, attributed primarily to Kolmogorov and Wiener, is mentioned and briefly described, but the bulk of the chapter is devoted to Wold's predictive decomposition theorem, and deductions from it made by the authors. By using the innovational representation of a nondeterministic stationary time series (that is, the output of minimum delay filter whose input is a white noise process), the authors set the stage for the succeeding chapters on predictive deconvolution. Here, we learn how predictive deconvolution is used to remove multiple reflections and reverberations from seismic traces. The wave propagation theory is the familiar one-dimensional, plane wave theory. It does not conform to the usual common depth point stacking procedures and does not always give good results. The newer slant stacking procedures are better and give more nearly the plane wave stacks for which predictive deconvolution is designed.

The book ends with a chapter on spectral estimation. Both the ARMA (auto regressive, moving average) and ME (maximum entropy) methods are described.

Robinson and Treitel have produced a useful, carefully written book for students, teachers and prospectors. Many of the subjects that they consider are important beyond the realm of seismic prospecting for oil, and their book should find appeal among all scientists and engineers interested in digital signal processing. □

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GFD for all levels

D. Anderson

Geophysical Fluid Dynamics. By J. Pedlosky. Pp.624. (Springer-Verlag: 1979.) DM 83.50, $43.90. *Geophysical Fluid Dynamics for Oceanographers.* By J.J. von Schwind. Pp.307. (Prentice-Hall: 1980.) £19.45, $29.95.

THE science of physical oceanography is in the process of emerging from the dark ages, a process begun some 40 years ago and greatly accelerated in the past decade. The companion subject of dynamical meteorology has had a similar history but is slightly further advanced. Together they constitute geophysical fluid dynamics (GFD), an important new discipline which is attracting interest because of its relevance to improving weather forecasts and understanding climate.

With the expansion of the subject has come a demand for textbooks. The two reviewed here are intended for new graduate students or advanced undergraduates, but the levels of the courses for which the texts are suitable are totally different. To the novice student embarking on one of the rather compressed courses which tend to be offered at many institutions, Pedlosky would be rather daunting. Von Schwind, however, is sufficiently elementary that it could be picked up and read profitably by any new student. The subjects discussed in Chapters 3 and 5 are important — wind-driven ocean circulation, the explanation of western boundary currents and some aspects of large-scale wave dynamics — though the presentation could have been markedly improved if some indication of modern thinking had been included (only three of the references are post 1970). Much of the remainder of the book is in the nature of support material. Chapter 2, by far the largest (over half the length of the book), deals with various aspects of basic fluid mechanics and geophysical fluid mechanics. Since many students enter

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oceanography with no prior knowledge of fluids, this can be quite useful even if much of it is readily available elsewhere.

Pedlosky on the other hand is suitable for more advanced graduate students and specialists. Almost everyone in this category is bound to find something of interest in this intensive book each time he opens it, be it for casual reading or reference. The scope of the book could have been more catholic but the treatment of his chosen material is generally very thorough, one notable exception being equatorial dynamics, an important aspect of modern thinking which gets only marginal coverage in the last chapter (and none at all from von Schwind). The mid-latitude quasi-geostrophic potential vorticity equation is well explained and there is a good section on instability theory. The presentation is mathematically, rather than physically orientated but Pedlosky is a text one can recommend to all serious students of GFD.

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Unmathematical air

John Green

Atmospheric Physics. By J. V. Iribarne and H.-R. Cho. Pp.212. (Reidel: 1980.) Dfl. 40, $15.95.

ANOTHER text that is said to require only elementary mathematics — in this case to understand the terrestrial atmosphere. I must confess a bias. To my mind *no* book on physics should use more complex mathematics than is needed to make its point. Conversely, no text should go out of its way to avoid some essential mathematics because the reader may not understand it. He had better be made aware of his deficiency and of the intellectual benefit that will result from being able to understand the mathematical part of an argument.

The authors identify the need for a survey of the physics of the terrestrial atmosphere, suitable for students having completed only one year of a course in general physics. They give a general description of the structure of the atmosphere, atmospheric chemistry, radiation, thermodynamics, cloud physics, and atmospheric electricity and dynamics. Each topic is treated sympathetically but briefly. At the end of each chapter there are questions and problems designed to aid the learning process, and hints on solutions at the end of the book. Diagrams are mostly well drawn but the writing is occasionally careless.

The philosophy of presenting material without recourse to more than elementary mathematics is admirable. Execution is less so. The student is given the appropriate formula with only the most general guidance to the most comprehensive monographs where, I believe, he will find the derivation very difficult to follow. More detailed reference to a text of intermediate standard would help. I think it is useful to comment that a stated formula contains some of the variables you would expect and has a plausible functional form, besides giving a detailed reference to a succinct derivation in the same notation.

This quibble apart, our poor, unfortunate, non-mathematical person will find this a useful, interesting and balanced survey of atmospheric physics.

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Star of the lecture theatre

Stephen P. Maran

Discovering the Universe. By Charles E. Long. Pp.511. (Harper & Row: 1980.) Pbk $16.50. *Dynamic Astronomy.* 3rd Edn. By Robert T. Dixon. Pp.523. (Prentice-Hall: 1980.) Pbk £11, $16.95. Teacher's manual also available.

AT A university in southern California, 500 undergraduates in a long, narrow auditorium observe a professor, dimly visible at centre stage, microphone around the neck, lecture on black holes, the Big Bang and life in space. At an East Coast university, a comparable throng files into two adjacent auditoria as the dividing wall between the rooms is electrically retracted; at the same time a large turntable at the front of one of the halls slowly rotates away the entomology instructor who has just finished a lecture. Into view spins the astronomer with demonstration table, microphone and piles of computer-graded examinations that are ready for return to the students. As the lecture begins and the lights fade away, a coloured photograph of a spiral galaxy appears overhead, projected in duplicate to provide identical side-by-side images for the benefit of those sitting at far left and far right in the wings of the assembly.

As the elementary, non-mathematical course in astronomy has become so popular in North America, perhaps 50 competing texts have appeared. The latest, *Discovering the Universe*, is alternately thoughtful and chatty, with many apropos anecdotes and numerous, seemingly-scrawled sketches that are as effective as the usual formal diagrams. At its best (taken on its own terms), *Discovering* introduces the unwary to general relativity with the aid of two inquisitive ants (Einstant and Newtant), or describes Mercury as seen by *Mariner 10*. In the latter case, only a few details are given, but much effort then follows in explaining what one might experience on a visit to the planet. The explanation is clear enough that the student will understand why each prospective phenomenon is anticipated. Marring the otherwise accurate text, however, are erroneous statements (each of which I have seen before) to the effect that *Uhuru* discovered Cygnus X-1 and that the optical pulsar in the Crab Nebula was discovered on the famous Lick Observatory rotating-shutter photographs. Those photos were obtained well after telegrams announced the discovery of the optical pulses at Steward Observatory, their confirmation at McDonald Observatory, and their identification with a specific star — the pulsar — at Kitt Peak National Observatory.

Dynamic Astronomy, here in a third edition, is distinguished by "flip pages", which bear in the margins sequences of diagrams that can be made to spring into life. Riffle them and you will observe Halley's Comet in retrograde motion, tail growing as it approaches the Sun, then shrinking as the comet hurtles back into the far reaches of the Solar System. Other sequences depict planetary, stellar and star cluster motions. Better yet, try turning the same pages slowly and you will find besides the diagrams a well-polished text, familiar by now to the students "in over two hundred colleges and universities". Yet improvements are still to be made. A section on "Observing Meteors" gives the wrong explanation for why more meteors are observed after midnight than before. Contrary to Fig.9-15, globular clusters are strongly concentrated to the centre of the Galaxy although they are more conspicuous in the halo. The sources of the gamma-ray bursts discovered by Klebesadel with data from the *Vela* satellites are confused with the very different "X-ray bursters" studied by Grindlay. Less significantly, although surprising in a mature text, the names of astronomers often are spelled wrong, including those of Olbers, Jastrow, Solomon and Maran. Somehow, when the reviewer's name is misspelled, it always is noticed. □

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The Institute of Biology has published a *Handbook for the Animal Licence Holder*, edited by H.V. Wyatt, intended particularly for newcomers to laboratory animal work. The *Handbook* is available from 41 Queen's Gate, London SW75HU, price £2.50 or £2 for five or more copies.

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Quantum mechanics for librarians

R. B. Jones

Simple Quantum Physics. By P. Landshoff and A. Metherell. Pp.177. (Cambridge University Press: 1980.) Hbk £12, $29.95; pbk £4.25, $9.95. *An Introduction to Quantum Physics.* By A. P. French and E. F. Taylor. Pp.670. (W.W. Norton/Nelson: 1979.) Hbk $17.95, £15.95; pbk $11.75, £8.95. *Principles of Quantum Mechanics.* By R. Shankar. Pp.612. (Plenum: 1980.) $29.50, £18.59.

AT THE end of any introductory quantum mechanics course, the lecturer sadly concludes that there is not yet a satisfactory textbook while the bemused undergraduate inscribes on the desk top "Schrodinger waives the rules — probably". The three books considered here do not solve the lecturer's problem although each has its merits. They are well produced with a few misprints and each is well provided with problems for which solutions are given at the end of the text. Apart from this they have little in common.

Simple Quantum Physics is based on lectures given at Cambridge University to mathematicians and physicists, and is suitable for a one-term introductory course. I enjoyed reading the book for its brevity and clarity. There is a very brief account of the basic ideas of wave mechanics in which the standard problems are treated up to the hydrogen atom without spin. The mathematical treatment is clear but so concise that only able students will follow what is going on. A short treatment of perturbation theory leads to the most distinctive feature of the book, an excellent introductory account of atomic absorption and stimulated emission and how these can be used to understand a laser. The final three chapters introduce solid-state quantum physics by way of band theory and the physics of real transistor devices. I will recommend this book to our college library and urge my students to read it; however I will use a more detailed text as the required reading in my own course of introductory quantum mechanics.

Out of the MIT Introductory Physics Series comes French and Taylor's *An Introduction to Quantum Physics*. Here the emphasis is on the experimental basis of quantum physics, the coverage ranging from the great historical experiments to modern laser spectroscopy. The basic approach is wave mechanical, although two chapters on photon polarization experiments point the way to the abstract notion of a state vector. All the basic problems up to the hydrogen atom are treated, and both spin and hydrogen fine structure are mentioned. Atomic radiation is discussed although no systematic perturbation theory is included. The mathematical treatment is unfortunately sparse. The student who uses this book will enjoy it, but he will have to do it all again. For that reason I cannot recommend it as a set text in British universities; however, in North America, where there is more time for an undergraduate degree, I would commend it highly for a one-term or one-semester course. Even in Britain many lecturers will want this book for its outstanding illustrations and examples, and libraries should have a copy for students.

Principles of Quantum Mechanics by Shankar is intended for a year-long first course in quantum mechanics. In contrast to the previous authors, Shankar introduces the subject in a loose axiomatic way using abstract linear spaces and the Dirac notation. The attempt to explain the subject at this level means that the book is very long, and even long-winded at times, without covering as much physics as one should expect in 600 pages or so. There are some compensations, however. There is a clear introduction to the Feynman path integral formalism and special attention is paid to the role of symmetries in quantum mechanics. All standard perturbation theory is covered, including fine structure effects in hydrogen. Angular momentum addition is treated and the concept of an irreducible tensor operator is introduced. Scattering theory, electromagnetic field quantization and even the Dirac equation are briefly discussed. This book has merits as a reference book, but I think its place is not in the lecture room. □

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Approaches to nuclear reactions

James Lowe

Introduction to Nuclear Reactions. By G.R. Satchler. Pp.316. (Macmillan, London/Wiley, New York: 1980.) £23, $59.95. *The Physics of Nuclear Reactions.* By W.M. Gibson. Pp.338. (Pergamon: 1980.) Hbk £19.50, $45; pbk £6.95, $16.75.

BOTH of the authors of these two volumes set out to provide a textbook on nuclear reactions, as opposed to static nuclear properties, for the undergraduate student. Usually, a single book covering the whole of nuclear physics provides adequate detail for an undergraduate course, and several such books are available. There are, in fact, some advantages in having the entire subject in one book — the unity of the subject renders the division into static properties and reactions arbitrary and sometimes confusing. However, the extra detail covered in a specialist book may well be useful to the more inquisitive student, especially if he intends to pursue the study of nuclear physics beyond undergraduate level.

Two books apparently aiming to teach much the same general topic at the same level could hardly be more different in their content and approach. Satchler's book, after an excellent introductory chapter, and an extensive section on basic scattering theory, covers in detail all the theories and models which have formed the basis of our understanding of nuclear reactions over the past 20 years. The treatment is authoritative; many of these models have been developed by Satchler himself. However, the approach is by no means historically orientated — topics of current interest, such as heavy-ion scattering and multi-step processes, are discussed and the book contains plenty of examples of the application of the methods described to "real live" data. The descriptive material is excellent, although curiously the only point at which it is a little misleading is the discussion of the concept of cross-sections, which is sometimes difficult to get across to students. The scope of the book is broad, both in the depth of treatment and in the wide range of topics covered. For example, even Glauber theory, which is useful at energies much higher than those usually encountered in nuclear reaction studies, is described briefly.

By contrast, Gibson's book, which is a new edition of his book *Nuclear Reactions*, provides a much more concise account of the basics of nuclear reaction theories. Unfortunately, several topics which are an essential part of most undergraduate courses are omitted; for example, there is no quantitative treatment of direct reactions. The discussion is clear throughout, and Gibson seems sometimes to assume a more elementary starting point for his readers than does Satchler. Gibson provides an excellent discussion of particle accelerators (though not of any other aspect of techniques — a curious selection), and also gives an account of the physical principles of fission and fusion reactors. His final chapters on nuclear forces and nuclear spin give good accounts of these topics, with a breadth of treatment beyond that directly relevant to nuclear reactions.

The choice between these books for the student must be based on their content. The student who intends to concentrate on pure nuclear physics will find Satchler's treatment valuable, and the content of this

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book will be useful well into the student's postgraduate years. In fact, the treatment is sufficiently extensive and not too elementary for many research physicists. For the student who intends to move towards applied nuclear physics or energy research, especially if involved in the physics of reactors, Gibson's book may be the more relevant. □

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Astrophysics

R.J. Tayler

The Physics of the Interstellar Medium. By J.E. Dyson and D.A. Williams. Pp.194. (Manchester University Press/Halsted: 1980.) £11.95, $24.95. *Radiative Processes in Astrophysics.* By G.B. Rybicki and A.P. Lightman. Pp.382. (Wiley-Interscience: 1979.) £16.95, $39.85.

ASTRONOMY, as well as being an exciting subject in its own right, gives a student an excellent opportunity to see how different branches of physics are brought together to solve a particular problem. For that reason, stellar structure, which involves atomic and nuclear physics, thermodynamics and statistical physics, electromagnetism, fluid dynamics and gravitation, is a popular option for physics undergraduates. The physics of the interstellar medium provides another excellent example of the interplay of different branches of physics and chemistry, such as the origin and transfer of radiation throughout the electromagnetic spectrum, hydrodynamics and magnetohydrodynamics and the physics and chemistry of small particles and molecules. Although it is a good subject for an undergraduate course, there is a lack of suitable textbooks. There is one outstanding book, *Physical Processes in the Interstellar Medium* by L. Spitzer (Wiley-Interscience, 1978), but this is far too advanced for the average or weak undergraduate.

The book by Dyson and Williams is written for the undergraduate audience and contains a range of topics which almost exactly matches the lecture course given to third-year undergraduates at Sussex. To some extent there is emphasis on the research interests of the two authors, interstellar hydrodynamics (Dyson) and interstellar grains and the formation and destruction of molecules (Williams), but overall it gives a good impression of the range of physical problems met in the interstellar medium. It is clearly written and contains a useful number of examples, and, as it uses SI units, it should be immediately intelligible, at least to UK undergraduates. It is a pleasure to be able to recommend to my students a book at a level suitable for all of them.

Rybicki and Lightman are also concerned with astronomy as physics but instead of dealing with a particular branch of astronomy they have considered all astrophysical applications of one branch of physics, the origin and transfer of radiation which is of particular importance in the interstellar medium. This is an outstanding book which has reached me when I am preparing an MSc course on the subject. It is making my task both simpler and more enjoyable, although I regret the need to translate its formulae from cgs to SI units.

Rybicki and Lightman have covered a wide range of topics, including the fundamentals of radiative transfer and a review of both electromagnetism and special relativity as well as discussing the various types of radiation from free particles (bremsstrahlung, synchrotron and inverse Compton radiation) and atomic and molecular structure and spectral lines. They have succeeded admirably in giving all the necessary information without too much unnecessary detail and the book contains many problems with worked solutions. The book should be of wide general interest to physicists as well as astronomers, since only the problems are specifically astronomical, and I hope that its title will not prevent them from recognizing its value. □

R.J. Tayler is Professor of Astronomy at the University of Sussex, managing editor of Monthly Notices of the Royal Astronomical Society *and author of three introductory astronomical textbooks.*

Facets of thermal physics

P. T. Landsberg

Principles of Thermodynamics. By J. A. Beattie and I. Oppenheim. Pp.329. (Elsevier Scientific: 1979.) Dfl. 98, $47.75. *Thermal Physics.* 2nd Edn. By C. Kittel and H. Kroemer. Pp.473. (W.H. Freeman: 1980.) $22.95, £11.30. Instructor's manual also available.

THE images of joint authors BO and KK (for short) transmitted by these two interesting and worthwhile books are rather different. BO (viewed as a unit) dresses in a formal dark suit, is reliable, a trifle old-fashioned and, because of a personal communication of 1911 acknowledged in the text (p.170), not a hot-headed youngster by any means. He has a good sense of the history of the subject: nineteenth century references are frequent, and only one or two literature citations from the 1970s occur — "new-fangled stuff compared with Clausius and Joule" one hears him say. He likes open systems and chemical potentials: van't Hoff's equilibrium box is not often discussed. Indeed there is an interest in fundamentals: Carathéodory's principle, the unattainability principle, Gibbs paradox are all considered though the commentaries and developments of these topics in the 1970s are not touched on. "Too new" said the author, replacing his pince-nez.

KK, on the other hand, wears a sports jacket and jeans, is most excited by newer developments, and readily departs from general principles to discuss effects and applications in this (largely rewritten) second edition: superconductors, solar constant, Overhauser effect, p-n junctions, heat pumps and particle–antiparticle equilibrium are all noted. Some of these interesting things are, however, in the problems. "Come on boys, this is fascinating stuff, you can do it", he says, while writing his concluding piece on negative temperatures. But there must always be a doubt on that point. An author may not even be able to solve his own problems if he has left the subject for a while.

If our authors should meet, one can imagine BO asking KK "Why is Nernst not mentioned or unattainability or Pfaffians?". "Well, there are special books for that" answers KK, puffing at his pipe; "Why don't you write one like that?". "I have" says BO.

Knowing some of the authors, the appearances inferred from their books are actually quite wrong. But the point is clear: thermal physics is a many-faceted subject and to get the full flavour it may not be enough to read one book. Even two, the above two, lucky though one is to have them, may not be enough. One would have to go elsewhere to learn about negative heat capacities, Saha's equation or black-hole thermodynamics.

This is not the place to make criticisms of details, but it may be of wider interest to note the following point (taken from my own book on the subject), which neither author stresses though they both deal with the zeroth law of thermodynamics. This law is $(A \equiv B, A \equiv C) \Rightarrow (B \equiv C)$. It states the transitivity of thermal equilibrium (denoted by $\equiv$), A, B and C being systems. Now comes a question which in its simplicity is suitable for the promotion board of the Scientific Civil Service: does the existence of a single temperature function follow, such that temperature equality and thermal equilibrium is the same? The answer is "No". This may be seen by interpreting "$\equiv$" as the condition for parallelism of lines, A, B, C in space. This requires *two* angles which specify the directions of the lines to be equal. Hence we have shown that cases exist in which a single function is inadequate. □

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Introductory . . .

W. Cochran

Elements of Solid State Physics. By M. N. Rudden and J. Wilson. Pp.186. (Wiley: 1980.) Hbk £14.50, $40; pbk £5.95, $17.

CHARLES Kittel maintains that "One test of a solid state course is that it should treat holes correctly". The book under review fails this test, since the authors cheerfully discuss the Fermi level in terms of the water level in a flask, a hole in terms of a bubble in the water, and that is almost their last word on the subject. However this is not meant to be a book for professional physicists, who will (perhaps) know that the wave vector of a hole state is *minus* the wave vector of the corresponding unoccupied electron state, a fact that cannot be of much interest to an engineer designing a large-scale integrated circuit. The authors are not lacking in ambition, for they have set out to write a textbook for the student who requires to be introduced to the Bohr model of the hydrogen atom on p.15, and who is yet presumed to be ready to understand the physics of the tunnel diode by p.153 — they are writing for

> the increasing number of students studying solid state physics at an introductory level, many of whom require only sufficient background to understand the operation of modern solid state electronic devices.

I began to read the book with some scepticism but I became increasingly impressed by the ability of the authors to convey ideas by means of arguments and analogies which, although they do not treat each concept "correctly", are almost always helpful and will not have to be consciously unlearnt by the student who goes on to study the subject more deeply. Once or twice I thought the authors were unhelpfully wrong, as when they write on p.87 that "for an intrinsic semiconductor at absolute zero the Fermi level coincides with the top of the valence band", then continue on p.88 with an unconvincing reason for its being "halfway between the two bands", and finally on the same page give a valid argument why it "can perhaps be best understood" to occupy the latter position.

All in all, the authors are to be congratulated on their success in writing a non-mathematical book on solid-state physics which is genuinely an introduction. Possibly students of electrical engineering need not know more than this about solid-state physics *per se* but, if they must, they will find several other books in the reading list, including inevitably Charles Kittel's *Introduction to Solid State Physics* (5th Edn, Wiley, 1979), which is close to the other end of the spectrum of introductions. □

W. Cochran is Professor of Natural Philosophy at the University of Edinburgh.

. . . and applied solid-state physics

J. C. Anderson

Introduction to Applied Solid State Physics: Topics in the Applications of Semiconductors, Superconductors, and the Nonlinear Optical Properties of Solids. By R. Dalven. Pp.330. (Plenum: 1980.) $27.50, £17.33.

THE philosophy of this book is that solid-state physics teaching, in physics courses, often lacks reality — a view with which I would not disagree.

The book is based on a lecture course, given by the author in the Department of Physics at the University of California, Berkeley. The course evidently set out to *survey* the *physics* involved (author's italics) in a variety of solid-state devices. To this end, the first two chapters rapidly review basic semiconductor physics and the p–n junction, giving liberal references to *Introduction to Solid State Physics* (5th Edn, Wiley, 1979) by Charles Kittel (who apparently encouraged the author to develop his course) and other relevant texts. The following chapters then deal with semiconductor devices such as the transistor (both bipolar and field effect), metal–semiconductor junctions and their applications such as Schottky diodes, and the present generation of devices available through MOS technology. A chapter devoted to other semiconductor devices is something of a rag-bag and rather unwisely attempts to encapsulate the physics of amorphous semiconductors in four pages. In this fast-moving field such an attempt is decidedly unwise. However the day is always saved at the end of each chapter by a suggested reading list and references to key papers, which should enable the reader to overcome any shortcomings of the exposition. The remaining three chapters deal with generators and detectors of electromagnetic radiation, superconductive devices and materials and, finally, nonlinear optical properties of solids.

Essentially, the book is offering itself to lecturers wishing to illuminate their solid-state physics teaching, and to those students who might have sufficient curiosity about the relevance of their solid-state physics course to go and read a book. In this context the book succeeds, as evidently did the author's lecture course. In a wider context it is also useful as a quick reference guide to a scientist or engineer wishing to remind himself, or learn for the first time, of the basic physics behind a device with which he has to deal. It successfully dispels, *inter alia,* the delusions that a squid is a kind of cuttle fish or that the Josephson junction is a crossroads near Bethlehem. □

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Electromagnetism, old and new

John Pain

Electricity and Magnetism. 3rd Edn. By W.J. Duffin. Pp.467. (McGraw-Hill: 1980.) Pbk £7.50, $18. *Fundamentals of the Theory of Electricity.* By I.E. Tamm. Pp.684. (Mir/Central Books, London: 1980.) £6.95.

FORTY years separate the first editions of these two books yet the essential contents are common to both. Most topics in physics advanced beyond all recognition during this particular period but the foundations of electricity and magnetism, as laid by the scientists of the nineteenth century, remain definitive.

Tamm is one of the great names of Russian physics; here an engaging slip in the biographical blurb awards him the Noble (*sic*) Prize. His book was first published in 1925 and ran to eight editions with only minor amendments before he died in 1971. His final preface acknowledged the need for a major revision but the ninth edition, published after his death, left the work unchanged. The book under review is a translation of that posthumous edition and the preserved style of the original anchors it firmly in the period of the inter-war years. This will commend it to a diminishing group of readers — those with a sense of history and a nostalgia for the European teaching tradition of that time. A lucid translation and good Romanian printing may encourage a wider audience but the theoretical bias, unrelieved by reference to practice, plus the use of an unfashionable system of units, will make no appeal to the physics undergraduate who has grown up in the world of radio, television, computers and communication satellites.

When Tamm first wrote this book radio was still in its infancy and he was under no pressure to include those experimental details which illustrate the extraordinary power of electromagnetic theory. An unrevised version of the same book is difficult to justify in 1981.

If Tamm's is the work of a past master, Duffin's is a model of what a modern textbook should be. The third edition deserves to be even more successful than the first two, for the author has expanded some of

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the more advanced topics and revised his presentation, helped by imaginative typography. This really is a book for all physics students (and course lecturers). The basic theory is there but solid physics flesh covers the mathematical bones without running to the fat of an over-written text. The book covers the first two years of an honours course in a British university and Duffin's technique is to take the early chapters slowly and to accelerate when the student is well grounded. His concern not to lose the reader is shown by commentaries and by revision summaries at the end of each chapter which bring the preceding pages into sharper focus. In addition there are nearly 300 problems and the answers are given to all of them with many useful hints.

There are few books which leave the impression that an outstanding lecture course has been transferred from black-board to page — this is one of them. □

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Learning soil physics

David L. Rimmer

Soil Physics. By T. J. Marshall and J. W. Holmes. Pp.345. (Cambridge University Press: 1980.) Hbk £33, $77.50; pbk £10.95, $21.95. *Applied Soil Physics: Soil Water and Temperature Applications.* By R. J. Hanks and G. L. Ashcroft. Pp.159. (Springer-Verlag: 1980.) DM 39.50, $22.20.

FOR some time an alternative has been required to the standard text on soil physics, *Soil Physics* by Baver *et al.* (4th Edn, Wiley, 1972), especially as it never appeared in paperback and eventually became too expensive for undergraduates to buy. The book by Marshall and Holmes is thus to be welcomed. In it they present a full account of the subject, concentrating in the first half on fundamental and theoretical aspects, especially of soil water, and in the later chapters dealing with the more field-orientated or management aspects, for example structural stability, compaction, tillage and irrigation.

The material is readable and attractively presented; tables and figures break up the pages, avoiding the problem of page after page of continuous text interspersed only by intimidating equations. Throughout there are many references; about 600 in total, these are brought together at the back of the book to provide a most useful reference list. There are, however, certain inconsistencies between chapters. For example, in Chapter 2 the total potential of soil water is defined in terms of pressure, osmotic and gravitational potentials only, where pressure potential is taken to cover matric potential under certain conditions. In Chapter 12 the total potential of water in plants is defined in terms of all four potentials separately. The choice of symbols and the use of SI units is generally good. Units for density however cause some problems; these appear in different places as kg m^{-3}, g cm^{-3} and Mg m^{-3}. Yet these are relatively minor criticisms of what is a very good book, which should become widely adopted.

Applied Soil Physics by Hanks and Ashcroft is different in many respects, although aimed at a similar readership (students of soil physics and scientists in related fields). First, its scope is more limited, as suggested by its sub-title *Soil Water and Temperature Applications.* Second, it specifically aims to show how the basic theoretical equations can be applied to field situations by means of numerous worked examples. These examples are backed up by sets of problems at the end of each unit (chapter). The aspects of soil water are dealt with in four units and soil temperature in a final unit. By working through the units the physical significance of the rather abstract theoretical relationships is made more apparent. The book is not strictly a textbook, but then it was not intended to be such (as evidenced by references to relevant textbooks at the end of each unit). What the book offers is an aid to teaching the subject. Its usefulness as such is somewhat diminished by the use of non-SI units, which may cause problems for students. But as a source of worked examples and relevant problems it will be of value to teachers of soil physics. □

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Pedology illustrated

Milo I. Harpstead

Soils: Their Formation, Classification and Distribution. By E.A. FitzPatrick. Pp.353. (Longman: 1980.) £25, $60.

SOIL science has attracted increased public awareness in recent years and has become the professional concern of scientists in a wide variety of fields. Soils can be lost by erosion and contaminated by pollutants, sometimes with disastrous consequences, and vary in their suitability to produce crops, support structures and accept waste products. Soils are being inventoried on a world-wide scale and soil survey reports are now consulted for purposes of land-use planning.

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normally taught in undergraduate courses. It is well illustrated with excellent photographs and drawings; of particular interest are the illustrations of landscape–soil relationships for several climatic regions of the world and the numerous photomicrographs from thin-sections showing the soil micromorphology resulting from the processes being described. In addition, there are 16 colour plates of soil profiles, representing major soil regions of the world, and a number of tables which represent an exhaustive compilation of information from international sources.

However, the book does have its faults. Mr FitzPatrick has reviewed the world literature on soils, but surprisingly he seems to have missed the point in his critical assessment of Hans Jenny's opinion about the factors of soil formation. Less importantly, he cites secondary sources for the theories of Davis and Penck on slope development, giving the impression that they were formulated several decades later than they were.

Many authors have developed their own nomenclature similar to that of Mr FitzPatrick; such terminology is usually short-lived and confusing if it does not reflect broad acceptance within the discipline. The author reminds us that soils form a continuum in space and time and that "this situation defies classification". He contends that for 80 years pedologists have tried unsuccessfully to construct a satisfactory hierarchical system of soil classification and this leads him to conclude that it simply cannot be done. The USDA uses the most elaborate hierarchical system in the world and so receives Mr FitzPatrick's severest criticism, an assessment which is contentious, to put it mildly.

This is a valuable book for pedologists, but has limited value for students who are attempting to master an established system of soil nomenclature and classification. □

Milo I. Harpstead is Professor of Soil Science at the University of Wisconsin, Stevens Point, and the author of Soil Science Simplified (*Iowa State University Press, 1980.*)

All round food production

J. C. Bowman

Agricultural Ecology: An Analysis of World Food Production Systems. By G. W. Cox and M. D. Atkins. Pp.721. (W. H. Freeman: 1979.) £15.70, $29.95.

A RIGOROUS analysis of world food production systems involves an understanding of many aspects of science and social science. One method of teaching agriculture is to consider the relevant disciplines of chemistry, biology, economics and so on, and then gradually to bring this knowledge together to show how it relates to food production systems adopted on farms. Undoubtedly this approach tests the patience of students and requires them to have faith that those aspects of science and social science which they study are relevant and will eventually interrelate to give a sound understanding of agriculture. Students can gain confidence and make a more purposeful approach to the choice of constituent subjects if they have an overview of agriculture from the start of their studies.

Cox and Atkins have made a brave and largely successful attempt to provide such an overview by considering agriculture as a group of ecosystems. Their book is in three parts. The first deals with the ecological and historical context of agriculture, the second with the dynamics of agroecosystems and the third with agriculture and the future. For a text with such a wide range of subjects which require integration and focus there is a big advantage in the easy and consistent style which the authors have achieved. There is a great deal of quantitative material which has been made comprehensible, even enjoyable.

It is perhaps inevitable that with only two authors some imbalance of subject matter has arisen. In particular there is considerable emphasis on agricultural pest problems — nearly half of Part Two is devoted to this topic — and there is much more attention given to crop rather than livestock production systems. This may represent, albeit inadvertently, the authors' view of the future importance of these subjects. A stronger element of the anthropological and sociological aspects of agriculture throughout the world, particularly in appreciating changes in agriculture in the developing world, would have been beneficial. Even so, the text gives a good account of the ecological approach to agriculture.

The book is profusely illustrated with photographs, diagrams and tables. Some of the diagrams have a minimum of explanation and in several cases students will need to return to the source for a full appreciation of what they portray. Each chapter has a good bibliography which draws heavily on American literature, and contains relatively little reference to publications after 1976 even though the book was not published until 1979.

The book should be valuable to all students of food production systems throughout their course. A text to be read and re-read as a student gains knowledge of the constituent parts of the subject. □

J. C. Bowman is Director of the Centre for Agricultural Strategy at the University of Reading.

Conserving resources

Ronald F. Hensler

Soil and Water Conservation (For Productivity and Environmental Protection). By F.R. Troeh, J.A. Hobbs and R.L. Donahue. Pp.718. (Prentice-Hall: 1980.) £16.85, $25.95.

THE first two chapters of this readable and well-edited book give an historical as well as a world-wide perspective on soil erosion. As well as providing students with a broad view of their subject, they should be required reading for those making decisions which in any way determine how land will be used, and thus to what degree that land will be subject to erosion by water and wind.

With the principles laid down, the authors go on to evaluate the parameters involved in water and wind erosion forces and soil-loss predictions. References here, as throughout the volume, are a useful combination of the key original concepts, recent research, and review articles.

Chapters 8, 9, 10 and 12 cover soil and plant management and conservation structure techniques (tillage, cropping systems, terraces, dams and so on) that can be employed to reduce erosion. Critical design requirements as well as discussions of the pros and cons of each technique are provided. The remainder of the book deals with soil surveys and land-use planning, mine and construction site reclamation, pasture, range and forest management, water conservation, drainage, irrigation, soil and water pollution, economics, United States soil and water conservation agencies, and conservation around the world.

Other features include a summary, (though not of the thought-provoking variety), and references at the end of each chapter. The illustrations are appropriate, and the subheadings have been well planned with each subsection able to stand by itself so that a teacher can assign portions of chapters without losing the integrity of the whole.

The major difference between this and other books on soil and water conservation is that most textbooks are engineering-orientated to meet the on-the-job needs of a practising soil conservationist. This book, then, is particularly welcome in filling a gap.

Soil and Water Conservation should certainly be considered for adoption by teachers of soil and water conservation courses. In addition, it will be a useful reference book for all agronomists, all politicians and all who make decisions that affect land use. I only hope such people read it. □

Ronald F. Hensler is Associate Professor of Soil Science at the University of Wisconsin, Stevens Point.

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BOOKS RECEIVED

Mathematics

BELLMAN, R. Analytic Number Theory: An Introduction. Mathematics Lecture Note Series, No.57. Pp.195. ISBN 0-8053-0360-X. (Benjamin/Cummings, Reading, Massachusetts: 1980.) $19.50.

FREEDMAN, H.I. Deterministic Mathematical Models in Population Ecology. Pp.254. ISBN 0-8247-6653-9. (Marcel Dekker: 1980.) SwFr. 64.

JONES, W.B. and THRON, W.J. Continued Fractions. Analytic Theory and Applications. Encyclopedia of Mathematics and its Applications, Vol.11. Pp.428. ISBN 0-201-13510-8. (Addison-Wesley: 1980.) $37.50.

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Astronomy

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DE JAGER, C. The Brightest Stars. Geophysics and Astrophysics Monographs. An International Series of Fundamental Textbooks, Vol.19. Pp.458. Hbk ISBN 90-277-1109-7; pbk ISBN 90-277-1110-0. (Reidel: 1980.) Hbk Dfl. 140, $73.50; pbk Dfl. 60, $31.50.

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STRUB, A.S. and IMARISIO, G. Hydrogen as an Energy Vector. Commission of the European Communities. Proceedings of the International Seminar held in Brussels, February 1980. Pp.703. ISBN 90-277-1124-0. (Reidel: 1980.) Dfl.95, $50.

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ROBERTS, C. Science, Animals and Evolution: Reflections on Some Unrealized Potentials of Biology and Medicine. Contributions in Philosophy, Number 14. Pp.221. ISBN 0-313-21479-4. (Greenwood, London: 1980.) £10.95.

ROY, A.K. and CLARK, J.H. Gene Regulation by Steroid Hormones. Pp. 316. ISBN 3-540-90464-6. (Springer-Verlag: 1980.) DM 84, $49.60.

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COHEN, P. Recently Discovered Systems of Enzyme Regulation by Reversible Phosphorylation: Molecular Aspects of Cellular Regulation, Vol.1. Pp.273. ISBN 0-444-80226-8. (Elsevier/North-Holland: 1980.) $63.50, Dfl.130.

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LEVI-MONTALCINI, R. Nerve Cells, Transmitters and Behaviour. Proceedings of a Study Week at the Pontifical Academy of Sciences, October 1978. Pp.679. ISBN 0-444-80243-6. (Elsevier/North-Holland: 1980.) $109.75, Dfl.225.

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Psychology

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